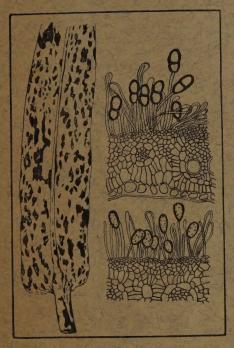
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# FACTORS AFFECTING VARIABILITY IN CEREAL RUST REACTIONS<sup>1</sup>.

## I. Variability caused by Temperature.

#### T. N. SHUKLA

(Accepted for Publication, October 6, 1953)

#### INTRODUCTION

Certain wheat varieties are stable in their reaction to races of stem rust in practically all environments, while other varieties are resistant to certain rust races in one environment but susceptible to the same races in another environment (Hart, 1949; Johnson, 1931). Since sensitivity to temperature, and possibly to light, is the most common cause of variability in several host-parasite complexes, temperature and light were investigated more thoroughly than other factors for the nine selected varieties of wheat inoculated with races 15 and 15B of stem rust.

#### EXPERIMENTAL

## (a) Effect of constant temperature and low light intensity.

Seven varieties of common wheats and two varieties of durums were planted at 75°F., and six sets of seedlings were inoculated with an isolate of race 15 and six sets with an isolate of race 15B. After 48 hours in moist chambers at 79°F., duplicate pots of each variety-race combination were placed at 80°-85°F., at 70°-75°F., and at 60°-65°F. Thus, although the seedlings were grown under uniform conditions and opportunities for infection were comparable at the time of inoculation and throughout the moist-chamber period, the stem rust races developed on each variety at three different temperatures. Light was approximately the same in the three sections when lightmeter readings were taken, usually three times each day. During this particular experiment light was rather poor, the intensity ranging generally between 1000 and 2000 foot candles.

Rapidity of rust development varied with temperature. The chlorotic flecks that indicate incipient rust lesions appeared in abundance on the seedlings at 80°-85°F. within six days after inoculation, and rust usually sporulated on susceptible varieties on the eighth day. At 70°-75°F., flecks also first appeared on the sixth day but were less abundant than at 80°-85°F. Uredia did not appear until the ninth day at 70°-75°F. At 60°-65°F., however, eight days elapsed before flecking and 10 days before rust sporulation on the same susceptible hosts. No difference was noticed in the rapidity with

<sup>1.</sup> The work was carried out at the University of Minnesota, U.S.A., under Drs. E. C. Stakman and Helen Hart. Sincere thanks are due to them.

which 15 and 15B developed, the two races being about equally sensitive to the temperatures used.

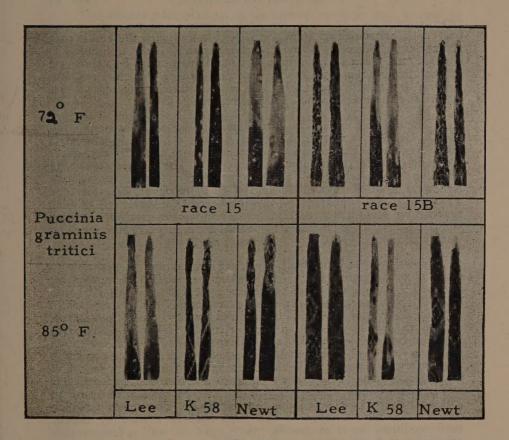
The common wheats Marquis and Mida and the durums Mindum and Stewart were relatively stable in rust reaction at the three temperatures and were susceptible to both stem rust isolates (Table 1). The only perceptible influence of temperature on the quality of rust infection was the slight variability within infection type 4. At 60°-65° and 70°-75°F. race 15 produced infections ranging from type 4— to 4 and race 15B produced types ranging from 4— to 4+. Better sporulation at 80°-85°F, resulted in an infection type 4 for race 15 and an infection type 4++ for race 15B and demonstrated the slightly greater virulence of 15B for these four varieties.

TABLE 1

Infection types produced by races 15 and 15B of Puccinia graminis tritici on seedlings of wheat varieties inoculated and incubated and held in a moist chamber at 79°F. for 48 hours, then transferred to different temperatures<sup>a</sup>

THE RESERVE OF THE PARTY OF THE		A STREET, LONDON	and the second
Rust race and	Infectio	n types <sup>b</sup> produce	d at
wheat variety	60°-65°F.	70°-75°F.	80°-85°F.
P. graminis tritici 15			
Lee Marquis Mida Mindum Stewart Frontana Newthatch Kenya 117A Kenya 58	O; to 1= 4- to 4 4- to 4 4- to 4 4- to 4 2 to 3cn O; to 1+ O; O;	O; to 1= 4— to 4 4— to 4 4— to 4 4— to 4 2 to 3cn O; to 1+ O; to 1= O; to 1=	O; to 1=  4  4  4  4  4  4  4  4 to 4+  4 to 4+
P. graminis tritici 15B			
Lee Marquis Mida Mindum Stewart Frontana Newthatch Kenya 117A° Kenya 58°	4- to 4+ 4- to 4+ 4- to 4+ 4- to 4+ 2+ to 3cn x+ O; O;	4 to 4+ 4- to 4+ 4- to 4+ 4- to 4+ 2 to 3cn 4- to 4+ O; O;	4 to 4++ 4++ 4++ 4++ 3 4++ 3cn 3cn

- a. Light intensity ranged from 1000 to 2000 foot candles.
- b. Infection types were classified according to Stakman et al. (1944). The notation on indicates an irregular chlorosis and necrosis of host tissues surrounding the stem rust lesions.
- c. A general leaf tip necrosis was present on the two Kenyas infected with 15B,



#### PLATE I

Fig. 1. Reaction of Lee, Kenya 58 and Newthatch wheats to races 15 and 15B at two different temperatures.

The reactions of Lee wheat also were relatively stable at the three temperatures, this variety being uniformly resistant to race 15 for which infection type ranged from O; to 1= and susceptible to race 15B as indicated by infection type 4 (Table 1; Plate 1, fig. 1). Again the high temperature (80°-85°F.) permitted better sporulation of race 15B than did the moderate or low temperature.

Frontana, with an intermediate reaction to the two rust isolates, was somewhat variable with temperature. Both rust races were equally pathogenic to the variety. The high temperature permitted sufficient sporulation for development of infection type 3 without noticeable chlorosis, but neither race 15 nor race 15B ever produced an infection type 4 at 80°-85°F, and the variety was only moderately susceptible. At the low and moderate temperatures the infection

type varied from 2 or 2+ to 3cn, and chlorosis and necrosis of the host cells accompained most of the rust lesions with the result that the variety appeared to be moderately resistant. If the uredium was situated on a green island of host tissue and surrounded by a chlorotic or necrotic ring the type 2 was recorded, but if the chlorosis and necrosis developed irregularly and were insufficient to reduce rust sporulation an infection type 3cn was recorded (Table 1).

The greatest variability in rust reaction was noted in Newthatch and the two Kenya wheats, K58 and K117A. These three wheats were susceptible to race 15 at high temperature and were extremely resistant at moderate and low temperatures. The rust infection type was 4 on Newthatch at 80°-85°F. and varied from 4 to 4+ on the two Kenya at 80°-85°F. At 70°-75° and 60°-65°F., however, infection type 0; or O; to 1+ appeared on these wheats (Table 1). A similar reversal in stem rust reaction with temperature was reported for another Kenya wheat infected by several different stem rust races by Darley and Hart (1944).

Newthatch and the two Kenyas likewise varied in reaction to race 15B with temperature. The Kenyas were moderately susceptible at  $80^\circ$ - $85^\circ$ F. and an infection type 3cn appeared on both K58 and K117A; but they were extremely resistant as  $70^\circ$ - $75^\circ$  and  $60^\circ$ - $65^\circ$ F. and race 15B never developed beyond the flecking stage. Newthatch, on the other hand, was very susceptible to race 15B at  $80^\circ$ - $85^\circ$ F., with infection type 4++; it also was susceptible at  $70^\circ$ - $75^\circ$ F., with an infection type 4+ to 4+; but the reaction was mesothetic at  $60^\circ$ - $65^\circ$ F. with infection type X+. The reaction of Newthatch to race 15B therefore changed at a lower temperature than that for race 15. It was not determined whether Newthatch was extremely resistant to 15B at temperatures still lower than  $60^\circ$ - $65^\circ$ F. The plants on which the infection type X appeared had been slightly shaded by nearby corn seedlings, so it is possible that light as well as temperature affected rust development.

Because four of the nine selected wheat varieties were more susceptible to race 15B at 80°-85F, than at 60°-65°F, and because of the indication that the critical temperature for a susceptible reaction might vary with the host-rust combination, other experiments were made at temperatures approximating 90°-95°F, atleast during day time. By chance, the experiments were made during bright, sunny weather when light intensities were two to four times those recorded for the first series of experiments.

# (b) Effect of high temperature and high light intensity.

Only three varieties of wheat were included in these experiments. Lee was chosen as the most susceptible variety and served as a control on the effectiveness of the inoculum of race 15B. Frontana was selected to find out if it might be completely susceptible to 15B at 90°-95°F. It had been only moderately susceptible to that rust race at 80°-85°F. The third variety used was Kenya 58 and it too had been only moderately susceptible to 15B at 80°-85°F.

The wheats were planted on six successive days and grown at 75°F. Seedlings also were inoculated on six successive days and as each lot was removed after 24 hours in the moist chamber it was placed beside the other lots on the bench in a greenhouse at approximately 92°F. during day time. For three weeks weather conditions were remarkably uniform, so that approximately the same quantities of rust and the same rust infection types developed on each variety in the six lots. One representative lot is presented in Table 2. The daytime temperature usually ranged between 90° and 96°F., although on two occasions 86° was recorded and on two other occasions temperature reached 98°. At night the temperatures dropped to 80°F. (Table 2a). Light intensity reached 5000-6000 foot candles on eight of the fourteen days for this representative run and on only one day was it as low as 1400 foot candles (Table 2a).

With the high daytime temperature (90°-96°F.) and the better light, rust developed faster than it had at 80°-85°F. and under low light intensity. Flecking was noted on the fifth day after inoculation, and uredia appeared on the sixth day.

In none of the six experimental runs were Frontana and K58 completely susceptible to race 15B. They were moderately susceptible as they had been at 80°-85°F. Rust infection type 3- to 3cn developed on K58 and the leaf tip necrosis again was observed on infected plants. On Frontana an infection type 3 occurred, but severe brown necrosis also developed around each rust lesion. In outward appearance the brown necrosis was similar to the browning that McFadden (1939) noted in mature plants of Hope and Hope derivatives infected with stem rust races 11,17, and 21, and it also was similar to the browning reaction that Hart and Allison (1933) reported in such wheat varieties as Acme, Arnautka, Einkorn and others on which races 11 and 34, and a few other races, developed at temperatures exceeding 83.3°F.

TABLE 2

Infection types produced by race 15B on Lee, Frontana and Kenya 58 in the greenhouse.

Wheat variety	Infection type <sup>a</sup>
Lee	4 to 4++
Frontana	3bn
Kenya 58 <sup>b</sup>	3- to 3cn.

a. In the other six sets similar infection types were observed. bn means brown necrosis. cn means chlorosis and necrosis accompanying uredia.

b. A general leaf tip necrosis also was present on infected K58 seedlings.

TABLE 2a

Average day temperature and maximum light intensity during the period of experiment summarized in table 2<sup>a</sup>

Consecutive days	Average daytime temperature in degrees F.	Maximum light intensity in foot candles
1 2 3 4 5 6 7 8 9 10 11 12 13 14	68-70 <sup>b</sup> 90 92 96 98 90 86 92 95 86 95 94 98 Final notes	 6000 6100 6000 5000 3800 1400 6000 5000 2400 3200 5000 5000

a. Average day temperature and maximum daily light intensity in the other six sets were almost the same. The night temperature was approximately 80°F.

b. Temperature in the incubator.

On Lee wheat at 90°-96°F, the infection types 4 to 4++ closely resembled the infetion types that had developed at 80-85°F. No modification of rust development was observed.

Apparently the critical temperature for a moderate susceptibility to race 15B in Frontana and Kenya 58 lies somewhere between 80° and 85°F., as observed in the first series of experiments; and a temperature above 90°F. does not result in complete susceptibility. Light intensity would seem to be a secondary factor to temperature in its effect on modifying rust reaction in these wheats except for the possibility of its influence on the development of the brown necrosis in Frontana.

Another type of experiment was made with race 15 at high temperature. Because rust is known to develop best on vigorous plants, an attempt was made to improve the vigour of the wheat seedlings by adding a complete nutrient solution to the plants while the rust was developing on them at the high temperature. The possibility that the combination of high temperature and an excess of nutrient might be more conducive to variability in rust reaction than the combination of high temperature and high light intensity had been, was investigated because earlier results with other hosts and other rust races had indicated that a high nutrient level had promoted rust sporulation and had reduced chlorosis of host cells, (Darley and Hart, 1944).

## (c) Effect of high temperature and nutrient:

Lee, Frontana, and Mida wheats, which had been resistant, moderately susceptible, and susceptible, respectively to race 15 at 80°-85°F. (Table 1), were planted in duplicate in a loam soil and grown at 75°F. The young seedlings were inoculated with race 15 and after 48 hours in the moist chamber at 75°F. the inoculated plants were transferred to a glass cabinet in which the temperatures varied between 89° and 93°F. One set of the varieties received 50 ml. nutrient solution daily in each pot; the control set received equivalent amount of tap water.

The nutrient solution used was similar to one recommended by Hoagland Arnon (1950) as a complete nutrient and it was prepared as follows:

Solution 1	ml. in a lite	er of solution
M NH <sub>4</sub> H <sub>2</sub> PO <sub>4</sub> (ammonium acid p	hosphate)	1
M KNO <sub>3</sub> (potassium nitrate		6
M Ca (NO <sub>3</sub> ) <sub>2</sub> (calcium nitrate)	•••	4
M MgSO <sub>4</sub> (magnesium sulpha	nte)	2
Solution 2.ª	gm. in a lite	er of water
H <sub>3</sub> BO <sub>3</sub> (boric acid)		2.86
MnCl <sub>2</sub> .4H <sub>2</sub> O (manganese chlorid	de)	1.81
ZnSO <sub>4</sub> .7H <sub>2</sub> O (zinc sulphate)		0.22
CuSO <sub>4</sub> .5H <sub>2</sub> O (copper sulphate)		0.08
(NH <sub>4</sub> ) Mo <sub>7</sub> O <sub>24</sub> .4H <sub>2</sub> O (ammonium m	olybdate)	0.02
Solution 3.		
Iron tartrate	0.05 per	cent solution.

Immediately before the nutrient solution was to be applied one ml. of solution 2 was added to each liter of solution 1. Twice each week one ml. of solution 3 was added.

Plants supplied with complete nutrient solution were healthier and more vigorous than those of the control. But the nutrient supply did not change the resistant reaction of Lee to a susceptible one nor the moderate susceptibility of Frontana to a complete susceptibility. The susceptible reaction of mida also was not changed. Sporulation of race 15 was improved slightly by the addition of nutrient. For example, on Lee wheat infection type 1+ was produced on plants with nutrient and type 1 on controls (Table 3). A similar variability was observed in Mida; infection types recorded were 4 to 4+ for plants in the nutrient series and 4—for its counterpart that received only water (Table 3).

a Ammonium molybdate was substituted for molybdic acid in solution no. 2.

#### TABLE 3

Infection types produced by race 15 on seedlings of three varieties of wheat with and without nutrient and kept at 89-93°F. after incubation in the moist chamber.

	Infection typ	Infection type produced			
Wheat variety	with nutrient	without nutrient			
Lee	1+	1			
Mida	4 to 4+	4-			
Frontanab	3bn	3-c			

- a. The average light intensity was 3200 foot candles.
- b. bn means brown necrosis surrounding uredia; c means chlorosis.

In Frontana also there was a slight variation in the infection type, type 3 with brown necrosis surrounding uredia occuring in plants with nutrient and type 3- with chlorosis in the controls (Table 3). The brown necrosis in this series of experiments was related to the nutrient supply rather than to the high temperature; it did not appear in the control Frontana plants that also had been at 89°-93°F. and under approximately 3000 foot candles. It was similar to the brown necrosis on Frontana inoculated with 15B and kept at high temperature and high light intensity (Table 2). In the work of Hart and Allison (1933) no effects of nutrients on the browning reation had been noted.

Daily temperatures over periods of two to three weeks during a normal growing season in the spring wheat area seldom are so uniform as were the greenhouse temperatures maintained for the foregoing experiments. Temperature trends often change every few days, that is, three to five days of very hot weather may be followed by one to several days that are cool and probably rainy. Or, perhaps five to seven cool, cloudly and rainy days precede a week of hot, sultry weather during which temperatures gradually rise to 85°, 90°F. or even higher at mid-day. While a wheat variety such as Kenya 58 was moderately susceptible to the rust if temperatures were high during the two weeks when rust was developing it was not known whether high temperatures were needed throughout the period or whether high temperatures during a part of the rust developmental period were sufficient to bring about the moderate susceptibility.

It also is usual in a normal growing season for temperatures twary greatly from mid-day to mid-night, the night temperature often being 20 degrees lower than the day temperature. While the Kenya wheats had been moderately susceptible at high temperatures in the foregoing experiments, there had been no night temperature lower

than 80°F. in these experiments and it was not know if the rust reaction of the Kenyas would vary with the day to night temperature fluctuations that are common in nature.

Therefore, experiments were made in which the inoculated plants were kept at high temperatures during the day but at low temperatures at night.

## (d) Effect of low night temperature.

In one experiment duplicate sets of seedlings of Lee, Kenya 58, Kenya 117A. Frontana, Mida, Marquis, Newthatch, Mindum, and Stewart wheats grown at 75°F. were inoculated separately with races 15 and 15B and incubated for 24 hours. Rust then developed on plants kept side by side in a greenhouse under indentical conditions. The greenhouse temperatures were high during the day and low at night. For ten days the daytime temperatures were 83°-85°F. and on two days the temperatures were 87° and 90°F.; the night temperatures for the most part were 62°-65°F. except for two days when 67°F. was the lowest night temperature (Table 4a). The maximum light intensity during the period of rust development was from 3500 to 5500 foot candles except for one day when it was only 2400 foot candles (Table 4a).

TABLE 4

Comparative infecton types produced on seedlings of nine wheat varieties by races 15 and 15B of Puccinia gramins tritici in the green house.

Variety .	Infection type produced by		
	Race 15	Race 15B	
Lee Kenya 58 <sup>a</sup> Kenya 117A <sup>a</sup> Frontana Mida Marquis Newthatch Mindum Stewart	O; and 1 to 1+ 3++ to 4- 3++ to 4- 3- to 3 3++ 3++ 3++ 3++ 3++ 3++	4 to 4+ 2++ 2++ 3- to 3 4 to 4++	

a. General leaf tip necrosis was associated with 15B infection.

TABLE 4a

Average daily temperature and maximum light intensity during the period of experiment summarized in Table 4.

Consecutive days	Average temp degrees		Maximum light intensity in foot candles
1 2 3 4 5 6 7 8 9 10 11 12 13 14	72a 83 85 85 90 85 87 85 83 83 83 83 85 Final notes	72a 65 62 63 63 65 65 65 65 67 67 67	3500 4500 4000 3500 3500 4500 4500 6000 5000 5500 2400 4500

a. Temperature in the incubator.

Chlorotic flecks appeared on the fifth day after inoculation, one day earlier than they had appeared in the previous experiment at 80°-85°F. when the light intensity had been only 1000 to 2000 foot candles. Uredia appeared on the sixth day, whereas eight days had elapsed before the rust began to sporulate in the earlier experiment. The better light undoubtedly was responsible for the more rapid rust development.

A comparison of table 4 with the last column of Table 1 reveals that both the races varied but slightly in the production of infection types on all the varieties in these two experiments. In general, rust development was slightly less on each variety than it had been in the experiments summarized in Table 1. For example, race 15 produced infection type 3++ on Mida, Marquis, Newthatch, Mindum, and Stewart as recorded in Table 4, but type 4 at  $80^{\circ}-85^{\circ}F$ . as shown in Table 1. Similarly race 15B produced infection type 4++ on the above five varieties at  $80^{\circ}-85^{\circ}F$ . (Table 1) but at high daytime temperature and low night temperature the infection type ranged from 4 to 4++ (Table 4). Again Frontana was moderately susceptible to both the races, the infection type being 3-to 3 (Table 4) whereas it had been type 3 at  $80^{\circ}-85^{\circ}F$ . (Table 1). As previously, Lee was resistant to race 15 and susceptible to 15B.

The two Kenyas were susceptible to race 15, infection type being 3++ to 4-; but they were moderately resistant to 15B, with

infection type 2++ (Table 4). Thus again the greater virulence of race 15 on these two Kenyas was noticeable as it had been at constant  $80^{\circ}-85^{\circ}F$ . (Table 1).

The Kenyas were moderately resistant (infection type 2++) to 15B in the present experiments while in the earlier experiment, reported in Table 1, they were moderately susceptible (infection type 3cn). The environments of the two experiments were similar in some respects but differed in others, and the differences probably account for the differences in rust reaction. During the daytime when temperatures in the present experiment were 83°-85°F. conditions were similar to the conditions of the first experiment when the temperatures were 80°-85°F.; and they favoured rust development. The night temperatures, however, differed in the two experiments; in the first one they were 80°-85°F, and thus favourable to a moderate susceptibility of the Kenya wheats; in the second one they were 62°-65°F., a temperature at which Kenyas had been very resistant to 15B. The high daytime temperatures probably allowed the rust to grow vegetatively and to sporulate but the low night temperatures retarded vegetative growth of the rust mycelium and rust sporulation. The antagonistic effects of these two temperatures probably tended to favour an intermediate reaction of the host and resulted in infection type 2++ and not type 3cn which had been produced at constant 80°-85°F. nor type O; which had occurred at 60°-65°F. (Table 1). Another factor, namely light probably influenced the rust reaction. Rodriguez (1945) observed that the chlorosis and necrosis of host cells proceeds rapidly at high light intensity when race 59 of P. graminis tritici infects Marquis wheat. Chlorosis and necrosis also occur very often in Kenya wheats and the greater rapidity of their development in the second experiment with 3500 to 5500 foot candles of light might account for the formation of a necrotic ring in a 2 infection type. In contrast to this the light intensity in the earliar experiment was 1000 to 2000 foot candles and the chlorosis and necrosis developed irregularly, so that no definite ring was formed.

These results emphasize again the great variability in rust reaction in the Kenya wheats and the great sensitivity of an intected Kenya to its environment during the period of rust development. Apparently several different infection types from O; to 3cn are possible, the one which finally develops depending on the particular post-inoculation environment.

#### DISCUSSION

The only known way to determine the reaction of a variety to a rust race is to inoculate the variety with that particular race and allow the rust to grow and sporulate. For successful infection, establishment, and development of the rust an optimum environment for certain period of time is required. This rust reaction is the indicator of the resistance or susceptibility possessed by the variety and is governed by (a) the genetic constitution of the host, (b) the genetic constitution of the rust, and (c) the interaction of these two factors and the environment. Three different rust reaction classes—resistant, mesothetic, and susceptible—are recognised. These classes are

determined by the types of infection produced by the rust race. An infection type is dependent on the vegetative growth of the mycelium, primary and secondary rust sporulation, and the amount of chlorosis and necrosis of host cells surrounding uredia.

Environment during the period of rust development may be of minor importance in determining the stem rust reactions of many wheat varieties, and the various factors of the environment may fluctuate within rather wide limits without altering the rust infection type appreciably. Varities like Mida, Marquis, Mindum, and Stewart were susceptible to races 15 and 15B under a temperature range of 60° to 98°F., either with low or high light intensities. Similarly, Lee was resistant to race 15 and susceptible to 15B in all the environments tried. Frontana also was moderately susceptible to both 15 and 15B under all conditions. This shows that these varities are stable in their rust reaction to 15 and 15B.

On the other hand, the development of stem rust in the Kenya wheats e.g. K117A and K58 is very dependent on the environment following inoculation and infection. At constant moderate and low temperatures with low light intensity, these two Kenya wheats were extremely resistant to 15B but at temperatures above 82°F., either with low or high light intensity, they behaved as moderately susceptible. In another environment where the day temperatures were high and the light conditions were good but the night temperatures were moderate to low, K117 and K58 both were moderately resistant to 15B. Thus it seems that the post—inoculation environmental limits, under which a particular infection type is produced by race 15B on these Kenyas, are narrow. Furthermore, it is evident that K117A and K58 can be resistant to 15B in one environment, moderately susceptible in another, and may have an intermediate reaction in a third one. The gradation between moderate susceptibility and extreme resistance depend on the particular environment under which the rust develops. On the basis of results it may be expected that K117A and K58 will be resistant in cool growing seasons and moderately resistant in warm growing seasons.

#### SUMMARY

The rust reactions of nine wheat varieties, Lee, Kenya 58, Kenya 117A, Frontana, Marquis, Mida, Newthatch, Mindum, and Stewart, to races 15 and 15B of *Puccinia graminis tritici* were studied under different environments in which temperature was the principal variable.

Stem rust reactions of certain wheat varieties were stable under a wide range of environmental conditions but varied considerably in two Kenya varieties, temperature playing the most important part. The two Kenya wheats reacted as moderately resistant at lower temperatures and moderately susceptible at higher temperatures with intermediate gradations between the two.

Application of complete nutrient solution did not produce any significant changes in rust reaction, but favoured a better plant development and better rust sporulation in almost all the cases.

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#### THREAD BLIGHT OF GINGER

#### N. V. SUNDARAM

(Accepted for publication October 6, 1953)

In Malabar district of Madras State ginger (Zingiber officinale Rosc.) is subject to infection by Pythium aphanidermatum (Edson) Fitzpatrick, P. myriotylum Dreschler and P. vexans de Bary, resulting in the rotting of the rhizome in storage as well as in the field and the wilting of the aerial shoots of the crop in the field (Thomas 1940). Rhizome rot of ginger caused by Pythium spp. has been reported from Bombay and Ceylon also (Uppal 1940, Park 1941). Sclerotium rolfsii Sacc. is also responsible for a similar disease causing shrinkage and rotting of the rhizome in storage and wilting of the aerial shoots in the field (Sundaram 1951). Phyllosticta zingiberi Ramak. is responsible for a leaf spot disease of the crop in the field (Ramakrishnan 1942).

In the month of August 1951 a new disease, namely, 'thread blight' of the leaves was observed at the Agricultural Research Station, Pattambi (Malabar). The initial symptoms of infection were noticed as small watersoaked lesions either on the margin or in other portions of the leaf. One or more lesions developed on the leaf involving a major portion of the leaf blade. The infected leaves became flacid and were observed to be hanging down or sometimes broke off from the sheath. Old and young leaves were found to be infected. Fine hyphal threads were observed to be spreading over the surface of the infected portion. In some cases small brown sclerotia were present on the lower surface of the leaves. On drying, the infected portion turned white and papery. This disease did not extend to the shoot but was confined to the leaf blade alone. Rapid spread of the infection was prevalent during the rainy months but with the advent of dry weather further spread was arrested.

#### MATERIALS AND METHODS

The fungus was brought into culture from surface sterilised tissues of leaves in the early stages of infection. Its growth was studied on different agar media prepared according to the standard methods (Riker and Riker 1935). The pathogenicity of the fungus was investigated by inoculating healthy plants grown specially for the purpose in pots and the inoculated plants were kept under bell jars to maintain high humidity for 96 hours. The inocula consisted of bits of young cultures containing mycelium and sclerotia of the fungus.

#### EXPERIMENTAL RESULTS

1. The Pathogen: The fungus grew rapidly on the media in the laboratory. Loose aerial mycelial growth of much branched

hyphæ 6-10 $\mu$  in thickness occurred on oats medium. In the course of 3-4 days chocolate brown sclerotia with a pubescent surface were formed. There was no development of the basidial stage on any of the media. The growth characters on four media are given in Table I.

TABLE I

Nature of Growth of the Fungus on Media

No.	Medium	Nature of growth	Time of formation of sclerotia	Size of Sclero- tia	Remarks
1	Potato dex- trose agar	Thick, whitish to buff, aerial	4th day	5-9 mm	Best growth
2	Onion agar	Thin and scanty, whitish, aerial	3rd day	4-9 mm	
3	Carrot agar	Thin and whitish	4th day	5-9 mm	
4	Oats agar	Thin and stringy, aerial, whitish	4th day	4-8 mm	

Though sclerotia were formed on onion agar early, the best growth as judged by the quantity of mycelium was on potato dextrose agar.

2. Pathogenicity of the fungus: In order to determine the parasitism of the isolate on ginger and the host range of the isolate inoculation experiments were conducted on healthy leaves of ginger and other plants. The results of inoculation tests are indicated in Table II and the symptoms have been shown in Plate I.

TABLE II

## Results of Inoculation Experiments

No.	Hosts	Part ino- culated	No. inocu- lated	No.	Description of infection
. 1	Ginger	Leaf	11	11	Watersoaked spots developed in 48 hours. The entire leaf rotted
2	Maranta arundinacea	—do—	8	8	in 3 days. Infection in 48 hours; entire leaf affected in
3	Canna indica	—do—	5	5	5 days. Spots in 48 hours. Entire blade affected in
4	Gossypium hirsutum	-do-	13	13	7 days.  Spots in 48 hours.  Whole leaf dried up
5	Groundnut	-do-	28	28	in 4 days. Growth in 72 hours. Entire leaflets rotted
6 .	Banana	-do-	2	2	and shed in 5 days.  Spots developed on the 4th day.
7	Citrus sinensis	do	7	6	Spots in 48 hours. Entire leaf involved in 4 days.
8	Setaria italica	-do-	6	6	Spots formed in 24 hours. The whole blade involved in 3
9	Barley	-do-	. 6	6	days. Spots in 48 hours; whole leaf affected in 4 days.

Suitable controls were kept and all of them were healthy throughout.

It is seen from the results of the inoculation experiments that the tungus has a wide host range and is capable of infecting both monocotyledonous and dicotyledonous plants. The limiting factor in the spread of infection was the relative humidity of the atmosphere. Saturated atmosphere favours rapid spread of infection but when the humidity goes down the spread of infection is arrested. This is in conformity with what was observed in nature where the disease was found only during the southwest monsoon.













PLATE I Symptoms of infection produced on leaves of :--

Maranta arundinacea.

Barley.

The hyphae of the fungus are found to spread over the surface of the inoculated portions as long strands. Some of these penetrated into the tissues through the stomata. Inside the tissues the hyphae spread intercellularly. Small irregular brown sclerotia are formed on the surface of the spots. These are much smaller than those formed on the agar media.

#### DISCUSSION

The pathogen closely resembles the one responsible for the banded leaf blight of Maranta arundinacea which was identified as Pellicularia filamentosa. Venkatarayan (1950) suggests that the fungus on M. arundinacea should be referred to some strain of S. rolfsii rather than Pellicularia. Comparison with several isolates of S. rolfsii available in this laboratory showed that the two fungi are entirely different and do not exhibit any smilarity. There are differences in the nature of the growth of these fungi on agar media. The type of sclerotium formed by S. rolfsii is quite distinct from the one formed by the fungus on ginger or Maranta.

#### CONTROL MEASURES

Diseases caused by *Pellicularia* are easily controlled by preventive spraying of copper fungicides before the disease occurs. On the Agricultural Research Station, Pattambi, Bordeaux mixture (5-5-50) was used for spraying a number of plots. This treatment gave a satisfactory control as judged by the incidence of infection. There was no spread of infection in the sprayed plots whereas in the control plots there was rapid spread of the leaf blight. Based on these results it is recommended that in places where this disease occurs on ginger the plants should be protected by spraying the leaves with Bordeaux mixture before the onset of heavy rain. But care should be taken to cover both the surfaces of the leaves with the fungicide.

My thanks are due to Sri T. S. Ramakrishnan, Government Mycologist, for his help in the investigation and in the preparation of the manuscript. My thanks are also due to Sri. K. SriRamachandran formerly Assistant in Mycology who has helped me in carrying out the experiments.

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# A CONTRIBUTION TO THE KNOWLEDGE OF UREDINEAE OF BIHAR

#### A. S. YADAV

(Accepted for publication October 9, 1953)

It is proposed to publish an account of the rusts which either have not been reported before on the hosts mentioned or have not at all been reported from Bihar and such additional information which has been gathered by the author during the course of his study.

1. Catenulopsora flacourtiae Mundkur & Thirumalachar, in Ann. Bot. 7: 27, 1943; B. B. Mundkur and M. J. Thirumalachar. Mycological Papers No. 16 Imp. Myc. Inst., Kew, Surrey, 1946.

Syn. Uredo uguressae Petch. Ann. Roy. Bot. Gdns. 1907; B. B. Mundkur. Sci. Mono. Coun. Agr. Res. India, No. 12, 1938.

Uredia epiphyllous, small, not coalescent, sub-epidermal, paraphysate, paraphyses slightly incurved, both peripheral and intermixed, uredospores borne singly on short pedicels with a single germ pore, 12-16×8-16  $\mu$ .

Telia hypophyllous, sub-epidermal, erumpent, paraphysate, spores forming long columns up to 24 spores in a chain, chains separate, spores more or less spherical, each jointed firmly at base to the spore below, without germ pores,  $14-16\times8~\mu$ , germinating in the sorus by the prolongation of the apical region into a long, four celled promycelium, sporidia up to 6-8  $\mu$  in diameter.

Hab. On the living leaves of Flacourtia ramontchi L'Herit., Parasnath Hills, Leg. A. S. Yadav (14-12-52). Both uredia and telia are present.

The rust has been reported on *Flacourtia sepiaria* Roxb. from Yeshwantpur (Mysore) and *F. sapida* Roxb. from Mahjgawn (U. P.). The host is a new record.

2. Uromyces leptodermus Syd., in Ann. Myc. 4: 430, 1906. Butler and Bisby, Sci. Mono. No. 1, Coun. Agr. Res. India, p. 82, 1930., M. J. Thirumulachar, Mycologia. 42: 2, 1950.

Syn. Uredo panici prostrati Sydow Ann. Myc. 4: 444, 1906.

Nigredo leptoderma Arth., N. Amer. Fl. 7: 224, 1912.

Hab. On the leaves of Brachiaria distachya Stapf., Patna., Leg. A. S. Yaday (2-12-52),

3. Uredo terminaliae P. Henn., in Hedwigia, 1896; M. K. Patel, V. P. Gokhle and N. B. Kulkarni, Indian Phytopath., 4: (1) 1951.

The uredial stage of the rusts has been reported from Mahableshwar (Bombay). The measurment of the uredospores in the present case (20-36  $\times$  12-20  $\mu$ ) differ slightly from the previous case (20-30  $\times$  15-21  $\mu$ ) recorded by Patel *et al.* 

Hab. On living leaves of *Terminalia chebula* Retz. Parasnath Hills, Leg. A. S. Yadav (14-12-52).

4. Scopella echinulata (Niessl) Mains in Ann. Myc., 37: 57-60, 1939; M. J. Thirumalachar, Mycologia 42: 227-230, 1950.

Hab. On the leaves of *Bassia latifolia*, Roxb., Sukhlal, Patna (3-5-50). A. S. Yadav (14-12-52). (only uredia and telia).

The collections made at different times showed that the telia appear in or after the month of March in Bihar. The uredial stage of the rust has been previously reported from Bihar (Kar).

5. Puccinia polliniae Barclay, in Mycologia 58: 243, 1889.

Telia chocolate brown crusts, sub-epidermal, spores yellowish brown, not constricted at septa, nor apically thickened, pedicel hyaline obliquely attached,  $28\text{-}32\times18\text{-}24~\mu$ .

Hab. on *Pollinia* sp. Parasnath Hills (4000 ft.), leg. A. S. Yadav (14-12-52). Only telia were present.

The rust was once reported from Simla (Barclay).

6. Puccinia suaveolens (Link) Rostrup; in Sydow H. & P. & Butler E. J. in Ann. Myc. 4: 439, 1906.

Hab. On the leaves of *Cnicus* spp., Bhagalpore, Leg. J. G. Srivastava. Only uredia were present. Butler has reported it from Pusa (Bihar) on *Cirsium arvense* Scop.

7. Puccinia heterospora Berk. and Curt. in Sydow H. & P. and Butler E. J. in Ann. Myc., 4: 432, 1906.

Hab. On living leaves of Sida veronicaefolia Lamk., Parasnath Hills, leg. A. S. Yadav (1-10-52.) Only telia were present.

The rust has been reported on Sida sp. from Bihar (Mitra). It has also been reported on Sida humilis Willd., S. spinosa L., S. mysorensis W & A., S. cordifolia L., by Butler form other parts of India. This host seems to be a new record for India.

8. Aecidium leucadinum Mitter., in Sydow and Mitter, Ann. Myc. 33: 55, 1935; B. B. Mundkur, Sci. Mono. Coun. Agr. Res. India, No. 12: 20, 1938; G. Watts Padwick, Mycological Papers No. 17, Imp. Myc. Inst. Kew, Surrey, 1946.

Pycnia minute, sub-epidermal, with ostiolar filaments. Aecia, sub-epidermal, erumpent, peridiate, spores in chains, 12-16 $\times$ 12-16  $\mu$ .

Hab. On leaves of Leucus montana Spreng., Parasnath Hills, Leg. A. S. Yadav (14-10-52).

The rust has also been recorded on the leaves of *Leucus mollissima* Wall from Nainital (Padwick, 1946).

9. Diorchidium levigatum Syd. & Butler, in Ann. Myc.

5: 485-595, 1907.

Uredia light brown, sub-epidermal, spores yellowish brown, echinulate, four equatorial germ pores. Telia dark brown, sub-epidermal, Spores slightly constricted at the septa, diorchnoid, thin walled,  $24-28\times16-24~\mu$ , pedicel hyaline.

Hab. On the living leaves of Oplismenus sp., Parasnath Hills. (4500 ft.), Leg. A. S. Yadav (14-12-52). Uredia and telia were present. The rust has once been reported on Oplismenus compositus Beauv., by Butler from Dehra Dun (U. P.) and also from Madras.

10. Puccinia pusilla Sydow, in Ann. Myc. 4: 435, 1906; Saccardo, Syll. Fung. 21: 693, 1912; Butler and Bisby, Sci. Mono. Coun. Agr. Res. India, No. 1: 73, 1931; G. Watts Padwick and Azmatulla Khan, Mycological Papers No. 10, Kew, Surrey, 1944.

Uredia sub-epidermal, cinnamon, spores ovoid to ovate,  $12\text{-}24\times20\text{-}24~\mu$ , with thick epispore, echinulate with four distinct equatorial germ pores, associated with club-shaped paraphyses. Telia black, small, sub-epidermal, spores slightly constricted at the septum, apically thickened (4  $\mu$ ),  $20\text{-}22\times28\text{-}34~\mu$ , pedicellate, pedicel hyaline attached either at the base or slightly oblique.

Hab. On living leaves of Bothriochloa pertusa (Fig. 1), Parasnath Hills (4000 ft.), Leg. A. S. Yadav (14-12-52) uredia and telia were present.

The rust was originally reported on Andropogon assimilis Steud. from Dehra Dun and also on Bothriochloa intermedia A. Camus. from Ranikhet (U. P.). This host is a new record for India.

11. Puccinia prainiana Barclay, in Sci. Mem. Med. Officers Army of India, 6: 65-69, 1891.

Syn. Caeoma smilacinis Barclay, in Sci. Mem. Med. Officers Army of India, 5: 37, 1890.

Telia light brown, in concentric rings, may be 1-5, separate, subepidermal, spores light brown, not constricted at the septum, slightly thickened at the apex, germinating immediately in the sorus forming a promycelium, upto 8  $\mu$  in thickness, 38-56×15-20  $\mu$ , pedicellate, pedicel hyaline, 40-48×4-6  $\mu$ .

Hab. On the living leaves of *Smilax macrophylla*, Roxb., Parasnath Hills. Leg. A. S. Yadav (14-12-52). Only telia are present. The rust has been reported on the same host from Mahableshwar (Bombay) and Madras.

12. Puccinia cynodontis Desm. in Sydow H. & P. and Butler E. J., Ann. Myc. 4: 436, 1906.

Hab. On the leaves of Cynodon dactylon Pers., Parasnath Hills, Leg. A. S. Yadav (14-12-52).

13. Puccinia cacao McAlp. in Saccardo, Syll. Fung. 21: 670, 1888; M. J. Thirumalachar (unpublished).

Pycnia sub-epidermal, pale yellow, older ones brownish, with ostiolar filaments, pycniospores upto 1.7 $\mu$  in diameter. Aecia sub-epidermal, peridiate, peridium one celled thick, in chains, 12-16x16-24 $\mu$ .

Hab. On the living leaves of *Hygrophila spinosa* T. Anders., (Fig. 2) Patna Leg. A. S. Yadav. (15.10.52). Only pycnia and aecia were present.

The pycnia appear usually in the month of October and the interesting feature is that these pycnia begin to die out and are followed not by aecia but another fresh crop of pycnia and it is in the month of December or last week of November that aecia begin to appear on the leaves. Cytological study of the older and new pycnia revealed that in the former the haploid mycelium bears sterile pycniophores, (Fig. 3) without bearing any pycniospores, whereas in the latter the same mycelium bears fertile pycniophores bearing pycniospores. It is only after the formation of the fertile pycnia that the aecia appear (this speaks of the significance of pycniospores in aecial formation. The detailed results will be published in a separate paper).







Fig. 2.

Fig. 1.—Puccinia pusilla Sydow on Bothriochloa pertusa, showing infection on leaves and leaf sheath.

Fig. 2.—Puccinia cacao McAlp. on Hygrophila spinosa T. Anders., showing infection on leaves.



Fig. 3.

Fig. 3.—Microphotograph of the pycnium of *Puccinia cacao* McAlp. showing sterile pycniophores with deposition of nectar on the ostiole.

Though the uredial and telial stages of the fungus have been reported from Pusa (Butler), Ranchi (Mitra) on Rottboellia compressa L., the pycnial and aecial stages were reported for the first time only from Patna by Thirumalachar (1952).

14. Cerotelium fici (Cast.) Arth., in Bull. Torrey. Bot. Club. 33: 30, 1906; M. J. Thirumalachar, D. V. Subba Rao and Ravindranath in Curr. Sci., 19, No. (1), 1950.

Syn. Uredo fici Cast. in Cook M. C., Grev. 5, 1876. Kuehneola fici (Cast.) Butler, in Ann. Myc. 12, 1914.

Hab. On living leaves of cultivated figs, Botanical garden, Science College, Patna, (8.11.52,) Leg. A. S. Yadav (14.11.52). Uredia and telia were present.

The rust seems to be perenial. The uredial stage occurs throughout the year on the leaves. Severe infection results in heavy defoliation of the shoot followed by rapid replacement. Weekly collections made from June 1952 revealed that the time of the appearance of the telia at Patna is after 1st week of November. Butler (1914) described the telial stage of the rust on Ficus glomerata Roxb, from Pusa. Thirumalachar et al (1950) described the telia of the rust on the cultivated figs for the first time in India from Banaras. This is the first record of the telial stage on cultivated figs from Bihar.

15. Ravenelia emblicae Syd., in Sydow H. and P., and Butler E. J., Ann. Myc. 10, 1912.

Hab. On the leaves of *Phyllanthus emblica* L., Parasnath Hills, Leg. A. S. Yadav (15.12.52).

16. Dasturella divina (Sydow) Mundkur and Kheswalla, in Mycologia, 35: 201-206, 1943. On living leaves of bamboo, Parasnath Hills, A. S. Yadav (14.12.52).

It has been found that the teliospores may either develop in seperate non-paraphysate telia or appear late in uredia. In the latter case the developing telial head totally replaces the uredospores within the paraphysate uredium. Thus the whole pustule appears like a paraphysate telium, a feature which has been overlooked by Mundkur and Kheswalla.

17. Puccinia bulbostylidicola Thirumalachar, in Mycologia, 39: 241, 1947.

Hab. On the leaves of *Bulbostylis barbata* Kunth., Patna, Leg. M. J. Thirmulachar, (25.10.52). Uredia and telia were present.

#### ACKNOWLEDGMENTS

The writer wishes to express his grateful thanks to Dr. M. J. Thirumalachar for kind help and encouragement. Thanks are also due to Shri J. G. Srivastava, Science College, Patna, for kindly identifying a number of hosts and Shri M. B. Sinha for preparing typed copies of the manuscript.

#### SUMMARY

A number of rusts collected from Bihar have been described and a short account of some of these having new hosts or showing some important features have been given with illustrations of those which seem to be interesting.

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# CONTRIBUTION TO KNOWLEDGE OF INDIAN ASPERGILLI

#### S. B. CHATTOPADHYAY AND C. DAS GUPTA

(Accepted for publication, October 14, 1953)

Mohanty (1948) after a detailed study of the different species of Aspergillus reported in India from time to time came to the conclusion that in India there are altogether 24 recognisable valid species of Aspergillus namely (1) Aspergillus amstelodami (Mang.) Thom & Raper, (2) Aspergillus candidus Link, (3) Aspergillus chevalieri (Mang.) Thom and Church, (4) Aspergillus fischeri Wehmer, (5) Aspergillus flavipes (Bain. and Sart.) Thom and Church, (6) Aspergillus flavus Link, (7) Aspergillus fumigatus Fr., (8) Aspergillus ipaponicus Saito, (9) Aspergillus luchuensis Inui, (10) Aspergillus nidulans (Eidam) Wint., (11) Aspergillus niger van Tieghem, (12) Aspergillus ochraceus Wilhelm, (13) Aspergillus oryzae (Ahlb.) Cohn, (14) Aspergillus phoenicis (Corda) Thom, (15) Aspergillus repens (Corda) De Bary, (16) Aspergillus rugulosus Thom and Raper, (17) Aspergillus sulphureus (Fr.) Thom and Church, (18) Aspergillus sydowi (Bain. & Sart.) Thom and Church, (19) Aspergillus tamarii Kita, (20) Aspergillus terreous Thom, (21) Aspergillus unguis (Emile-Weil and Gaudin) Thom and Raper, (22) Aspergillus ustus (Bain.) Thom and Church, (23) Aspergillus variecolor (Berk. and Br.) Thom and Raper and (24) Aspergillus versicolor (Vuill) Tiraboschi.

Quite recently from the soil of the paddy fields at the State Agricultural Farm, Chinsurah, West Bengal, India, 27 isolates of Aspergillus were obtained, which on detailed study were found to represent 9 (nine) species, namely (1) Aspergillus awamorii Nakazawa, (2) Aspergillus clavatus Desm., (3) Aspergillus flavus Link, (4) Aspergillus fumigatus Fr., (5) Aspergillus oryzae (Ahlb.) Cohn, (6) Aspergillus nidulans (Eidam) Wint, (7) Aspergillus terreus Thom, (8) Aspergillus versicolor (Vuill.) Tiraboschi and (9) Aspergillus violaceo-fuscus Gasperini. Of these Aspergillus awamarii Nakazawa, Aspergillus clavatus Desm. and Aspergillus violaceo-fuscus Gasperini are new records for India.

A short description of these three species is given below, as they are being reported for the first time in India.

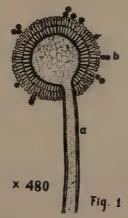
The representative cultures have been maintained in the culture collection of Mycology Section, State Agricultural Research Institute, Government of West Bengal, Tollygunge, Calcutta. They have also been deposited in the Indian Type Culture Collection, Divison of Mycology and Plant Pathology, Indian Agricultural Research Institute, New Delhi.

During the course of this study, the key for identification suggested by Thom and Raper (1946) was followed. The cultures were

grown in Czapek's soluion agar at a temperature of 23°c-25°c as recommended by them.

#### DESCRIPTION OF THE SPECIES

(i) Aspergillus awamari Nakazawa (in Inst. of Govt's. Res. Formosa, Rept. Vol. 1, 1907 and Vol. 2, 1912) Colonies in Czapek's solution agar spreading, growing very rapidly with abundant submerged mycelium, aerial hyphae scanty with large number of conidial



a. conidiophore of Aspergillus awamori Nakazawa showing stalk, vescicle and sterigmata. b. condia.

heads. Heads uniformly distributed throughout the surface. When fully grown, the colonies have a granulated appearance with wrinkling surface at the central region. Hyphae white in colour. Colonies when fully developed, blackish brown to deep chocolate in colour, substratum white. Conidiophores 1 to 1.3 m.m.  $\times 11.1-18.5\mu$  in breadth, averaging 1.2 m.m.  $\times 14.8\mu$  in breadth; wall smooth, colourless; foot cell prominent. Vescicle globose, large,  $37.5-55.1\mu$  in diameter, averaging  $48.1\mu$  in diameter. Sterigmata in two series, primary sterigmata  $4.8-7.4\mu\times3.2-3.4\mu$  averaging  $9.3\mu\times3.5\mu$ ; secondary sterigmata  $4.8-7.4\mu\times3.2-3.4\mu$  averaging  $9.6\mu\times3.3\mu$ , Conidia globose to subglobose; coloured, olivaceous to brown;  $3.2-4.8\mu$  in diameter in long axis averaging  $3.6\mu$ ; thickwalled, outer wall rough and spinulose.

Culture no A. 9 of Mycology Section, State Agricultural Research Institute, Tollygunge, Calcutta. Isolated from soil samples collected at 3" level from the paddy fields at the State Agricultural Farm, Chinsurah, West Bengal, by the senior author in June, 1950.

(ii) Aspergillus clavatus Desm. [in Ann, Sci. Nat. Bot. (2) 2:71, 1834]

Colonies in Czapek's solution agar growing moderately rapidly forming a thick coarse velvety mycelial mat; when fully developed,



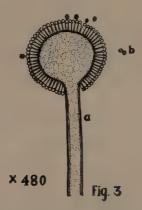
a. Conidiophore of A. clavatus Desm. showing stalk, vescicle and sterigmata. b. conidia.

with conspicuous radial furrows, bearing abundant conidiophores, with blue green heads arranged in very well defined zones. Mycelium white in colour. Conidial heads radiating, large, clavate, splitting into parts with age. Conidiopheres 1 m.m. or more in length  $\times 11.3-14.4\mu$  in diameter, thinwalled, smooth, colourless, gradually enlarging to form a clavate vescicle. The fertile area of the vescicle  $83-130\mu$  long  $\times 37-48\mu$  broad averaging  $111\mu\times 40\mu$ . Sterigmata in one series, much variable in size,  $6.4-9.3\mu\times 3.2-3.5\mu$  averaging  $7.4\mu\times 3.4\mu$  near the apix,  $3.5-4\mu\times3.0\mu$  averaging  $3.6\mu\times3.0\mu$  at the base. Conidia subglobose to elliptical, light green in colour, 3.2-4.0\mu in long axis, averaging  $3.6\mu$ , smooth.

Culture no. A.11 of culture collection of Mycology Section, State Agricultural Research Institute, Tollygunge, Calcutta. Isolated from soil samples collected at 3" level from the paddy fields at the State Agricultural Farm, Chinsurah, West Bengal by the senior author in September, 1950. This particular strain has smaller sized vescicle as compared with the strain obtained from culture collection of Indian Agricultural Research Institute, New Delhi by courtesy of Dr. R. S. Vasudeva, Head of the Division of Mycology and Plant Pathology, Indian Agricultural Research Institute, New Delhi.

(iii) Aspergillus violaceo-fuscus Gasperini (in Atti Soc. Toscana Sci. Nat. Pisa. Mem. 8, fasc. 2, 1887).

Colonies upon Czapek's solution agar growing moderately rapidly forming a thin mycelial mat producing abundant heads which are uniformly scattered throughout the entire surface. Colonies, when mature, purple brown to violet in colour, substratum colourless on the reverse. Conidial heads loosely radiating, purple brown to violet in colour. Conidiophores colourless, smoothwalled, 1 m.m. to 1.6 m.m.



a. Conidiophore of A. violaceo-fuscus Gasperini showing stalk, vescicle and sterigmata. b. conidia.

 $long \times 13-15\mu$  in diameter; foot cell present. Vescicle globose  $44.4-55.5\mu$  in diameter averaging  $51.8\mu$ , granulated within, fertile over the entire area. Sterigmata in one series only, loosely arranged.  $6.2-7.5\mu$   $long \times 3.4-3.7\mu$  broad, averaging  $7.3\times 3.6\mu$ . Conidia elliptical distinctly granulated, roughened, violet in colour when mature;  $4.0-48\mu$  in diameter in long axis,  $2.4-3.6\mu$  in short axis, averaging  $4.4\times 3.2\mu$ .

Culture no. A.27 of culture collection, Mycology Section, State Agricultural Research Institute, Tollygunge, Calcutta. Isolated from soil samples collected at top layer of the paddy fields at the State Agricultural Farm, Chinsurah, West Bengal by the junior author in May. 1952.

#### SUMMARY

A detailed description of the three species namely Aspergillus awamari Nakazawa, Aspergillus clavatus Desm. and Aspergillus violaceo-fuscus Gasperini recorded for the first time in India is given herewith. Including these, the number of recognisable Indian species is now 27.

State Agricultural Research Institute, Government of West Bengal, Tollygunge, Calcutta.

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# HELMINTHOSPORIUM DISEASE OF RICE—II. SOURCE AND DEVELOPMENT OF SEEDLING INFECTION

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#### INTRODUCTION

Helminthosporium oryzae Breda de Haan (Cochliobolus miyabeanus (Ito et Kurib.) Drechsler ex. Dastur) infects rice in all stages of its growth from germination to maturity. The infection taking place at germination which may be termed as seedling blight, forms the subject of the present study. The disease appears in germinating seedlings as small spots in the leaf sheaths and leaves. When such spots are numerous the seedling dies due to the infection. Seedling death also takes place without the appearance of any spots, when it just turns brown and collapses with frequently a profuse mycelial growth covering it.

Severe epiphytotics of Helminthosporium disease of rice have been recorded in India only at the flowering stage of rice varieties, though seedling blight has been reported from Sind (Uppal, 1929; Hingorani and Prasad, 1951). Seedling blight has been known to be exceptionally severe in Philippines (Ocfemia, 1924a) and Puerto Rico (Tucker, 1927).

Generally rice season extends from June to December except in those areas where a second crop is raised. In the absence of the crop after December, the appearance of primary seedling infection when seed beds are sown in June-July would depend upon (i) the fungus remaining viable during this period and (ii) its pathogenicity under the conditions prevailing in the sowing season. These two aspects were investigated in the present study.

#### REVIEW OF LITERATURE

(a) Viability of the fungus: Nishikado and Miyake (1922) reported that good growth was obtained from a culture of Helminthosporium oryzae two years and seven months old. In infected leaves also (presumably maintained in a dry state in the laboratory) Nishikado (1929) found that the fungus was viable for two years and seven months. Suzuki (1930) reported that the pathogen could remain viable inside the seeds for four years. According to Ito (1932), the conidia and hyphae of the fungus survived for 2 and 3 years respectively, when stored "indoors". Nishikado, Hirata and Higuti (1938) reported that the pathogen survived for 34 months at 0°, 5°, 15°, and 20° C and for 28-29 months at 30°C but the fungus was not viable for more than five months at 35°C. Page et al (1947) found that the

viability of conidia was best retained at low temperatures and that relative humidity of storage had more influence on longevity than temperature alone. For example at 20 percent relative humidity the conidia survived for six months at 31°C, whereas at the same temperature and over 95 percent relative humidity they lost their viability within a month of storage. They concluded that the conidia would not survive for long in the warm moist climates of the rice growing regions.

(b) Development of seedling infection: According to Ocfemia (1924b) the soil temperature at the time of germination of rice influenced the extent of seedling infection. At 16°C the infection was 100 percent, but at 24°C it came down to 10 percent and was totally absent above 30°C. Thomas (1941) showed that at 15°C the percentage of infected seedlings and death was 60 and 42 respectively, but at 29°C, they came down to 38.6 and 12 respectively. Padmanabhan et al (1948) reported that both pre-and-post-emergence blight was noticed under laboratory conditions, though no seedling blight was observed when infected seeds were sown in the field.

#### MATERIALS AND METHODS

A. Viability studies: The viability of the fungus in infected plant materials like leaves, nodes, inter-nodes and in soil was determined for the period December-July with a view to find out the sources of infection to the seedlings at the sowing season (June-July). The investigations given below were mostly carried out during 1949-50 season, except where specifically mentioned otherwise. Infected leaves, nodes and inter-nodes of plants were collected from early maturing varities in November and stored in grease paper bags in the laboratory. The viability of the fungus in these was compared with the viability of the fungus in infected stubbles of the crop remaining in the field after harvest by monthly isolation from these materials. Isolations were made from 4—6 samples of each material in a month.

The viability of the seed borne pathogen was determined with seeds of five varieties of rice, viz., T. 1145, T. 1242, T. 812, T. 412 and T. 90. Four oz. samples of seeds were stored in desiccators in 10%. 25%, 50%, 75%, 90% relative humidities over appropriate sulphuric acid dilutions and also over water (100% relative humidity). The seeds were stored in April and samples of 100 seeds were drawn in June and July for isolation of internally borne fungi.

The viability of the fungus was determined in sterilized soil in Erlenmeyer flasks and in unsterilized soil in pots. Both were inoculated with the fungus and the former was stored in the laboratory while the latter was kept exposed to natural conditions. Subsequently after completion of the studies on the effect of soil temperature on the development of seedling infection mentioned below, the viability of the fungus in soil was tested with reference to its pathogenecity on germinating seedlings at favourable ranges of temperature during May and June. Two sets of Erlenmeyer flasks of 250 c.c. capacity containing 50 gm. of soil, 5g m. of rice powder and 10 ml, water were

prepared and one set was sterilized at 20 lb. pressure for two hours. Both the sterilized and unsterilized lots were inoculated in April with a culture of the fungus and stored under room temperature. Seeds of a susceptible variety, D.C.A. 2, were treated with hot water (52°C for 15 minutes) to free them from internally borne infection and were sown in the sterilized and unsterilized soil contained in the flasks in May and June. After 48 hours when germination had started, the seeds along with the contents of each flask were transferred to sterilized soil in cigarette tins and maintained at 18-22°C in a wooden ice chamber. This was charged with 20 lb. of ice every 24 hours to maintain the required range of temperature.

The viability of the fungus in sterilized oatmeal agar slants maintained at 10°C and at room temperature (25-35°C) was also determined by monthly insolations.

B. Development of seedling infection: The development of seedling infection was studied with reference to the soil temperature at the time of germination. The experiments were conducted in February-March 1950 when the range of room temperature was 28-30°C and in January 1951 when the range of room temperature was 20-26°C. Helminthosporium oryzae was grown in 1000 ml. Erlenmeyer flasks in sterilized soil-rice medium (finely sieved soil 200 gm., rice meal 20 gm. and water 25 ml., sterilized for two hours at 20 lb. pressure) for 21 days at room temperature. At the end of the incubation period the mycelial mat was about 5-6 mm. thick. It was thoroughly mixed up with the medium and added on to the top of sterilized soil in cigarette tins and seeds of ten rice varieties were sown on the inoculated soil. A set of uninoculated sterilized soil sown with the same ten varieties, served as control. After sowing, daily observations were taken on seedling emergence and infection. In the first test the sowing was done on 22-2-1950 and daily observation on the progress of germination and development of infection was taken till 8-3-1950.

The second test was carried out in precisely the same manner between the 4th and 20th January 1951 with 7 varieties included previously and three new varieties.

C. Aeroscope studies: In order to determine whether the conidia of Helminthosporium oryzae are aerially borne and whether they are present over rice field at the time of sowing, greased slides were exposed in aeroscopes over rice fields in the months of April to July.

Six aeroscopes were installed at six locations in the Farm on the bunds of rice fields. The height of the aeroscopes was 2 ft. 6 in. Greased slides were exposed for 24 hours inside the aeroscopes at weekly intervals. The number of conidia of Helminthosporium orvzae found in the slides was counted.

#### RESULTS

A. Viability test: The data on the viability of the fungus in leaves, nodes, internodes, stubbles, in soil and in culture are presented in table 1.

TABLE 1

Showing the viability of Helminthosporium oryzae in plant parts stored in laboratory and in field, in soil and in culture during December to July (viability is indicated as +)

July	+ +							+ + +	+
radA VaM	+	++						+	+
Jan. Feb. Mar.	+	+		+	+			+	+
Feb.	+	+	+	+	+	+	+	+	+
	+	+	+	+	+	+	+	+	+
Dec.	+	+	+	+	+	+	+	+	+
Material	Infected leaf stored in laboratory	—op—	Infected node stored in laboratory	Stubble stored in laboratory	_op	Stubble in field	Unsterilized soil inoculated	Sterilized soil inoculated	Culture at 10°C
Variety	T.642	T.897	Benibhog	op	DCA.12	T. 642			
	A. Plant Parts						B. Soil		C. Culture

The fungus was found to remain viable throughout the period of test in culture, in sterilized soil and in infected leaves stored dry in paper bags. The fungus was also found to remain viable at 90 percent relative humidity and below in the seeds of all the five varieties upto the end of June. In July the fungus was viable in two of the varieties T.1145 and T.1242, over 90 percent relative humidity and in all the varieties at lower relative humidity percentages. The fungus was not viable beyond March and beyond April in stubbles stored in the field and in laboratory, respectively.

Out of the four to six samples of each plant material from which isolations were made, the number of successful isolations of Helminthosporium oryzae dropped down from month to month. Side by side, the number of isolations yielding ordinary moulds like species of Aspergillus and Penicillium increased, till finally, the loss of viability was marked by the plant materials yielding the moulds only in the isolations.

In the viability studies with sterilized and unsterilized soil in flasks, seedling blight developed in May (100 percent) and in June (30 percent) in the sterilized soil only. No infection was seen in the unsterilized soil (Table 2).

TABLE 2 Viability of Helminthosporium oryzae in inoculated soil tested by its pathogenicity to cause seedling blight (under  $18-22^{\circ}C$ )

		Percentage of seedling blight					
Variety 	Month	Sterilized soil inoculated	Unsterilized soil inoculated	Sterilized soil uninoculated			
DCA.2	May	100	nil	nil			
	June	<b>30</b>	nil	nil			

The fungus grew very vigorously in both the sterilized and unsterilized soil for the first three to four days after inoculation of the flask. Subsequently, the fungus continued to grow vigorously in sterilized soil but in unsterilized soil a considerable number of moulds soon filled up the flasks. After germination of the seeds it was observed that the seedlings in the unsterilized inoculated soil were very vigorous, green and tall compared to those in the sterilized inoculated soil. Even the seedlings which were free from infection in sterilized soil were short and stunted with small pale yellow leaves.

B. Development of seedling infection: In the first test when the temperature range was  $28-30^{\circ}\text{C}$ , only one case of seedling death was recorded. No leaf and stem lesions were observed. In

the second test when the temperature range was  $20-26^{\circ}C$ , leaf and stem lesions were observed in eight of the varieties, though seedling mortality was noticed in only 2 varieties. The control series also showed infection in two of the eight varieties. Since the seeds were not treated with hot water, infection in the control series might have been caused by the internal seed borne infection. It was noticed in both the tests that the fungus was actually growing on the surface of the soil and though the seeds were germinating on the mycelial mat, the fungus failed to produce any general seedling infection in the first test. The relevant data are presented in Table 3.

TABLE 3

Percentage of seedling mortality and leaf and stem lesion in 13
varieties of rice grown on inoculated soil at two different
ranges of temperatures.

	Teste	Tested in FebMar. 1950			Tested in January 1951				
Variety	Average room temperature 28 - 30°C					Average room temperature 20 — 26°C			
	Inoculated		Inoculated Control		Inoculated		Control		
	A	В	A	В	A	В	A	В	
T. 1145		•••				•••	•••		
N. 136		***	•••	***	•••	***	•••	•••	
Pb. 9	•••	***	***	460	4 .	16	•••	***	
T. 812		***	***	***		8	***	4	
PTB. 10	***	• • •	***	***	•••	8	•••	***	
AKP. 1	***		1		***	4	• • •	•••	
PT.B. 11	***	***	*** / "	***	20	4	4	4	
SLO. 5	4	*** '	***	• • •					
AKP. 2	•••	***	***	***					
T. 1242	***		***	***					
DCA. 2					***	24	• • •	•••	
T. 141					•••	16		***	
DCA. 12					***	4	***	•••	

- A. Percentage of seedling mortality.
- B. Percentage of leaf and stem lesions.

In addition to the experiments carried out with the object of testing the effect of temperature on seedling infection, experimental evidence which has a direct bearing on the point under discussion was also obtained from certain other investigations. A soil inoculation experiment was carried out during 25-9-1950 and 21-10-1950 in sterilized and unsterilized soil with a number of organisms isolated from spotted paddy grains, one of which was Helminthosporium oryzae. The variety used in the test was Benibhog, a susceptible

variety of short duration. The seeds were treated with hot water to free them from internally borne infection and sown side by side with untreated seeds in sterilized and unsterilized soil in small pots. No difference was observed in seedling emergence between the inoculated and the uninoculated series and no infection of the seedlings after emergence was noticed in any of the treatments. The range of room temperature during the experiment was  $29-33^{\circ}C$ .

Seedling blight resulting in death of seedling was observed in an isolated plant in an experiment on seed treatment carried out in November 1949. Helminthosporium oryzae was one of the fungi isolated from the dying plant. The range of soil temperature during this period was 19°C - 23°C. The isolate obtained from the plant was used in an inoculation experiment in pots in the third week of January 1950, when the range of soil temperature was 18 - 20°C. Seedling blight were observed in the inoculated series. The varieties Ptb. 10 and T. 412 were sown in this experiment on both of which the seedling blight occurred. Seedling blight has also been noticed in second crop seedlings during December-January in the fields in the years 1949-50, 1950-51, 1951-52 and 1952-53.

C. Aeroscope studies:—The number of conidia of Helminthosporium oryzae trapped in the slides exposed in aeroscopes over rice field in each week in the months, April, May, June, July in the years 1950, 1951 & 1952 are given in Table 4.

From the table 4 it may be seen that the conidia of *Helminthosporium oryzae* occur over rice fields in the months of April-July.

#### DISCUSSION

Helminthosporium disease of rice is seen in seedling leaves in July a few days after germination. A few isolated spots are seen in some of the beds under susceptible varieties. On the other hand, in the Philippines and Puerto Rico the disease is reported to cause severe seed-bed losses due to pre- and post-emergence blight. In India seedling blight has been noticed in the second crop season only in sowings made during November-January.

The experimental evidence adduced in the present paper that seed-borne *Helminthosporium oryzae* causes seedling infection only under cooler temperatures of 26°C and below (confirming in this respect the findings of Ocfemia in the Philippines and Thomas in Madras) explains the absence of primary seed-borne seedling infection in the main sowing season for rice in the plains of India when the soil temperature is always above 30°C.

When the possibility of seed-borne primary infection is precluded, the infection on seedling leaves seen in July was considered to be secondary in origin and aerially borne. This presumption was substantiated by the evidence from aeroscope study in which aerially borne conidia were seen to occur over rice fields during the sowing season and after. Thus aerially borne conidia causing secondary infection are responsible for the continuation of the disease cycle from season to season. The source of origin of these conidia is being studied.

TABLE 4

Showing the number of conidia of Helminthosporium oryzae trapped in six aeroscopic slides in different weeks.

	Total	31	27	rO	
	5th	:	m	nil	
July	4th	nil	Н	N	
	3rd	00	. 4	2	_
	2nd	00	nil		
	1st	15	liu 61	1 nil	
	1st 2nd 3rd 4th 5th Total	20	43	9	
	5th	:	:	:	
June	4th	5	4	nil	
, n	3rd		4	2 4 nil nil	_
	2nd	Ħ	12	4	_
	1st	4 11	23	N	
	Total	77	16	75	
	5th	30	70	H	
May	4th	9 4 30	3 nil	13	
4.	3rd	0	c	53	
	2nd	18 16	4	2 53	
	lst		<b>'4</b> '	9	
	Total	149	-	82	
	5th	;	-	9	
April	4th	26	:	00	
V	3rd	43	:	36	
	2nd	24	:	13   19   36	
	1st	56	:	13	
	I car	1950	1951	1952	

The seed borne fungus found viable in the seed presumably loses its viability in the soil at the sowing season. The fungus does not remain viable even a month in unsterilized soil whereas it continues to survive in sterilized soil for a number of months and retains its pathogenicity also.

## SUMMARY

The viability of *Helminthosporium oryzae* in seeds, leaves, nodes and inter-nodes, in stubbles left in the field after harvest and in soil was studied from December till July *i.e.*, from the begining of the harvest in one season till the sowing period of the next. It was found that the pathogen remains viable only in the seed during this period under natural conditions.

The seed-borne pathogen, however, fails to cause primary infection on seedlings at the time of germination at temperatures of 28°C and above.

Aeroscope studies have shown that the conidia of *Helminthosporium oryzae* occur over rice fields in the months of April to July, *i.e.*, during the sowing season and earlier.

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## BROWN-ROT OF MESTA (HIBISCUS CANNABINUS LINN)

## T. GHOSH AND K. V. GEORGE

(Accepted for publication, October 25, 1953)

Hibiscus cannabinus Linn is extensively cultivated for its fibre in the Vizag districts of Madras and Hyderabad. There are five varieties comprising eight distinct types which are all grown in India. It is also grown in Persia, Russia, Cuba, Central America. In U. S. A. it is known as 'Kenaf'. (Kundu: Business Week, 1951. Annual Number, Madras).

After the partition of India, due to shortage of jute in India, mesta, which is considered as the nearest substitute for jute, was introduced in many areas where jute could not be grown. As a result the area under mesta is on the increase.

Since 1950 it has been observed that the mesta crop in Bengal suffers from a serious fungal disease which usually starts from the end of July, attains maximum severity during August and continues till September. The authors have not come across any reference to any such similar disease in mesta.

Symptoms: The disease is caused by a fungus which attacks the young apical leaves, stipules, petioles, leaf-buds and the young stem towards the apex. Usually the attack starts on young tissues in general; the first visible symptoms appear on the young unfolded leaves and stipules; the anthocyanin pigment around the affected area becomes darker; the affected tissues turn brown to blackish, crinkle and finally disintegrate. The fungus also attacks the petiole forming dark necrotic areas eventually killing the leaf.

On the stem the first visible symptom is the formation of one or more irregular, or more often, fusiform brownish black streaks along the length of the stem, which are usually confined within 8 inches to 10 inches from the growing apex. The fungus enters the stem directly and once it reaches the vascular bundles, extends up and down the xylem tissue, the vascular tissue turns brown and gradually disintegrates forming internal cavities (Plate I, D). Meanwhile on the surface of the stem the brownish black streaks gradually extend in area (1.4 to 2 cm.) and with the disintegration of internal vascular tissue the peripheral tissues shrink and form the characteristic depressions (Plate I, A). These depressions in the beginning are covered with a powdery pinkish mass of spores. As the rotting proceeds, the peripheral tissues rupture resulting in semi-open cavities.

The necrosis is sometimes limited to the tender tissues of the leaf buds and stipules and does not reach the vessels, either because of partial resistance of the host or due to unfavourable climatic conditions. In such cases the stem does not break but turns into a withered stick (Plate I, C).



PLATE I Brown-rot of Mesta (Hibiscus Cannabinus Linn)

- Showing characteristic depression.
  Showing Sporodochia in culture with setae.
  Showing infected shoots.
  T. S. of stem showing disintegration of Xylem-tissue.

Etiology: In the affected tissues, in the early stages, mycelium may be seen but it is scanty, more so in later stages. The mycelium is hyaline, septate and measure 2.0 to  $2.6\mu$ . The fertile layer is composed of simple sporophores which are aggregated together forming erumpent, discoid sessile and regular sporodochia (Fig. 1.). The sporophores measure 26 to  $40\mu$ . The spores are borne singly (Fig. 2), oblongellipsoid, muticate, unicellular, thin-walled, hyaline but appearing pinkish in mass. The spores are mostly  $13.5\mu \times 5.0\mu$ . They range from 5.98 to  $15.6\mu \times 3.4$  to  $5.2\mu$ . Sporodochia and spore-formation are rather restricted to the early stages so that in advanced necrotic tissues spores may not be found.



Fig. 1.—Showing sporodochium ×500 Fig. 2.—Showing conidiophore and attachment of spore ×500

In culture, on Potato Dextrose Agar, the fungus forms a cottony white mycelium which as it grows turns pinkish buff due to the formation of abundant spores. As in nature, spores are formed at the tips of closely aggregated sporophores giving rise to a pinkish buff powdery mass. Dark brown sporodochia are produced in culture on Potato Dextrose agar. In each sporodochium, setae 6 to 14 in number,  $34.5\mu$  to  $115.0\mu$  long, are irregularly distributed (Plate I, B). The setae are brown, thick walled, 3 to 4 septate, straight or crooked, not very broad at the base and tapering towards the tip. It is important that setae are found only in artificial culture and not on the host.

Pathogenicity: Preliminary infection tests show that the fungus is an active parasite capable of causing infection in healthy unwounded tissues. Detailed experiment is in progress and will be reported later on.

Resistance: Of the several types of Hibiscus cannabinus Linn (received from South India, Madhya Pradesh, Indian Agricultural Research Institute, Uttar Pradesh, Russia, America and China) grown in this Institute, none has been found to be completely resistant to this disease; however, the 'American Commercial', an improved strain of U. S. A., is comparatively more resistant.

Identification: The fungus agrees in most part the description of Saccardo for the genus Volutella Tode, except for one character of the sporodochia which describes them as 'margine cilata'. For other characters it resembles Volutella. Hence, the pathogen is tentatively placed in the genus Volutella Tode. Further work is in progress, Specific identification is awaited.

## SUMMARY

In Bengal, mesta (Hibiscus cannabinus) has been found to suffer from a hitherto undiagnosed disease caused by a fungus tentatively identified as a species of the genus Volutella. The fungus attacks stipules, young leaves and apical parts of the stem causing 'brown-rot'. Attacked leaf-buds and leaves become necrotic and fall off. The necrosis in stem travels deep inside and may cause the stem-break. So far only one strain of H. cannabinus, the American Commercial has proved to be fairly resistant. Majority or all the indigenous types are more or less susceptible to this disease.

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# FACTORS INFLUENCING BACTERIAL SOFT ROT OF POTATOES

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## INTRODUCTION

The development of bacterial soft rot of potatoes in storage and transit is largely a matter of temperature and other environmental factors, as also conditions prior to marketing that predispose tubers to rot (Nielson, 1946; Dewey and Barger, 1948). Except for Hollis and Goss (1950), who used an authentic culture of Erwinia carotovora, all other workers have apparently conducted investigations with unidentified organisms. Brierley (1928) has, however, shown that there are at least four species of bacteria, namely, Erwinia carotovora (Jones) Holland, Erwinia atroseptica (van Hall) Jennison, Erwinia aroideae (Townsend) Holland and Bacillus mesentericus (Flugge) Migula which cause soft rot of potatoes in pure culture. In the present investigation, all the four organisms were employed for a comparative study to determine the effect of predisposing and postinoculation factors on tuber rot incited by them. It may, however, be mentioned that, although the senior author has made a large number of isolations from rotted potatoes during the last five years, there has been no evidence of the occurrence of Erwinia atroseptica in this country whereas the other three bacteria are frequently obtained.

## MATERIAL AND METHOD

Authentic cultures of Erwinia carotovora, E. aroideae, E. atroseptica and Bacillus mesentericus\* were obtained from the Indian Type Culture Collection.

Inoculations were made by Jones' method (1901) which may be briefly described as follows. The whole uninjured tubers were thoroughly washed in water and immersed in 95 per cent ethyl alcohol for 2-3 minutes to drive out air from pores and crevices. They were then transferred to a solution of 1: 1000 mercuric chloride for half an hour to sterilize the outer surface and finally washed them in several changes of sterile water. The tubers were then kept in sterilized moist chambers and inoculated with a 24-hour old culture of each isolate by pricking them with a sterile needle soaked in bacterial suspension. Inoculated tubers were incubated at  $27\pm1^{\circ}\text{C}$ . for 48 hours, after which, lesion volumes in cubic centimeters were calculated

<sup>\*</sup>According to Bergey's manual (1948, p 709), the European Bacillus mesentericus is only a stage of growth of Bacillus subtilis.

by applying the formula  $\pi r^2 d$ , as suggested by Hollis and Goss (1950). This method of calculation was followed throughout the present study.

Partial reduction in oxygen tension in desiccators was created by keeping a solution consisting of 50 cc. each of 25 per cent KOH and 25 per cent pyrogallic acid in the lower compartments. Different humidities were obtained by adding varying proportions of sulphuric acid and water (Buxton and Mellanby, 1934).

## EXPERIMENTAL RESULTS

Predisposing factors:—Potato tubers were thoroughly washed in water and surface-sterilised with 0.1 per cent solution of mercuric chloride for 15 minutes. They were then distributed in six lots of 15 tubers each and subjected to the following treatments: (i) presoaking in water; (ii) prechilling (2°C.); (iii) exposure to high temperature  $(35^{\circ}\text{C.})$ ; (iv) storage under reduced oxygen supply; (v) solar irradiation (air temperature  $40^{\circ}$ C.); (vi) control. The tubers were exposed to the first four treatments for 2, 5, and 7 days and to solar irradiation for one hour only. The control lot was kept at rcom temperature (25°-30°C.) for different lengths of time and inoculated with the four organisms. Some uninoculated tubers, which were given the same treatment as the inoculated ones, were also kept to serve as a check for outside infection. Data are given in Table 1. Each value represents the average calculated cylinder volume for five rot lesions on five potato tubers, expressed in cubic centimeters. Since time of holding had no appreciable effect on the development of tuber rot, amount of rot developed after 7 days of exposure is only given in case of the first four treatments.

TABLE 1

Effect of different predisposing factors on tuber rot

_	Average amount of rot in cubic centimeters						
Treatment	E. carotovora	E. aroideae	E. atroseptica	B. mesentericus			
Presoaking Reduced oxygen High temperature Prechilling Control I Solar irradiation Control II	5.7 6.0 4.6 3.8 1.7 3.3 2.3	8.9 8.0 6.0 5.3 2.0 2.6 2.1	7.1 5.5 3.9 5.3 0.7 3.5 3.3	6.0 2.7 2.4 2.4 0.9 2.0 1.5			

N.B.—Control I for the first four treatments and Control II for solar irradiation.

The results show that there is marked relationship between pretreatment of potatoes and subsequent tuber rot development. In general, treated potatoes show greater amount of rot as compared to controls. Presoaking and storage under reduced oxygen show the maximum effect in predisposing tubers to rot, whereas prechilling and exposure to high temperature are not so effective. Exposure of tubers to solar irradiation for one hour also predisposes them to rot.

Bennett (1946) observed that soft rot infection was mostly associated with immature tubers. Potatoes were, therefore, harvested from 28-2-51 to 19-4-51 at an interval of about 10 days, inoculated individually with the four organisms, and incubated at  $27\pm1^{\circ}$ C. for 48 hours. Controls were maintained by pricking the fully matured potatoes, which were harvested on 19-4-51, with a sterile needle. The results are given in Table 2.

TABLE 2

Effect of Maturity of Potatoes on tuber rot

	Average amount of rot in cubic centimeters*						
Harvest dates	E. carotovora	E. aroideae	E. atroseptica	B. mesentericus			
28-2-51	2.6	2.6	3.4	1.5			
10-3-51	2.0	2.6	3.4	1.4			
20-3-51	1.9	2.1	2.2	1.5			
30-3-51	1.8	2.2	2.4	1.4			
9-4-51	1.5	2.2	2.0	1.1			
19-4-51							
(fully matured)	1.4	2.0	1.8	1.2			

<sup>\*</sup>Average of five rot lesions on five potato tubers.

It is observed that, with the maturity of potatoes, intensity of rot tends to decrease. The results, however, appear to be marked in case of potatoes harvested in April and February. All the controls remained healthy.

Post-inoculation factors:—The effect of temperature and humidity on bacterial soft rot was determined by inoculating tubers and incubating them at different temperatures varying from 4° to 35°C. and humidities from 10 to 100 per cent. Variation in temperature and humidity during the experimental period was 1°C. and 2 per cent respectively. Ten tubers were inoculated with each organism at each temperature and humidity and the volume of rot was calculated after 48 hours incubation. Uninoculated controls were maintained in each case.

TABLE 3 Effect of temperature and humidity on tuber rot

Temperature		Percent—humidity					
(oC.)	10	35	65	90	100		
		E. carotor	vora				
4 20 25 30 35	0.0 0.8 1.1 S** S	0.0 1.6 2.3 0.4 S	0.1 2.3 7.6 0.9 0.5	0.2 5.0 8.1 1.2 1.6	0.2* 5.7 8.6 1.1 1.5		
		E. aroide	ae				
4 20 25 30 35	0.0 0.0 S S S	0.0 0.0 S S S	0.0 0.2 0.4 3.1 1.2	0.0 0.3 3.3 3.9 1.4	0.0 0.6 1.4 6.0 2.9		
		E. atrosept	tica				
4 20 25 30 35	0.0 0.2 0.1 S	0.0 0.3 0.1 S	0.2 1.9 0.7 0.2 0.2	0.2 2.2 1.5 0.8 0.3	0.3 2.1 2.7 0.6 0.7		
B. mesentericus							
20 25 30 35	0.0 0.2 0.2 0.2 S	0.0 0.3 0.4 0.3 1.0	0.6 0.3 0.4 0.6 0.9	0.8 0.4 0.5 0.8 2.1	0.6 0.6 0.6 0.9 1.8		

<sup>\*</sup>Average amount of rot in cubic centimeters.

The data indicate that Erwinia carotovora and E. atroseptica are more actively parasitic at low temperatures, whereas E. aroideae and Bacillus mesentericus at high temperatures. E. aroideae does not produce decay at 4°C. even when the relative humidity is 100 per cent. Relative humidity below 35 per cent is not conducive to development of soft rot.

<sup>\*\*</sup>S=shrivelling of the tubers, but no rot.

## DISCUSSION

These studies have shown that tuber rot that develops in storage and transit is mainly governed by four factors, namely, maturity of potatoes, moisture, extremes of temperature and poor aeration. In India, however, tubers are not washed before marketing or storage. Temperature and aeration are, therefore, of considerable importance in this country and require particular attention, since it has been shown that exposure of tubers to solar irradiation of even one hour predisposes them to rot. It is also indicated that immature potatoes are more susceptible to bacterial rot than fully matured ones and this point should be borne in mind at the time of harvest.

Considering the temperature requirements of the four organisms in relation to parasitic attack, they fall into two groups: (i) E. carotovora and E. atroseptica with low temperature requirements and are therefore important during cold storage operations; and (ii) E, aroideae and Bacillus mesentericus, which are destructive at higher temperatures, and therefore more harmful during transit, particularly in India where temperature in wagons rises considerably due to respiration. Furthermore, it it indicated that low humidity is unfavourable for the advance of rot. These findings generally agree with those of Brielery (1928). The fact that none of these organisms causes rot at 4°C. and a relative humidity of 35 per cent may be successfully utilized for keeping soft/rot under check in cold storage.

The writers wish to express their grateful thanks to Dr. R. S. Vasudeva, Head of the Division of Mycology and Plant Pathology, for his keen interest taken in this work and for critically going through the manuscript.

## SUMMARY

- 1. Presoaking in water, storage under reduced oxygen tension. exposure to extremes of temperature and solar irradiation predis-pose tubers to bacterial soft rot.
- 2. It has been shown that intensity of rot tends to decrease with maturity of potatoes.
- 3. E. carotovora and E. atrosepatica are more destructive at low temperatures and E. aroideae and Bacillus mesentericus during transit when the temperatures tend to be high. All the four organisms, however, fail to produce rot at 4°C. with relative humidity up to 35 per cent.

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## A PROBABLE STRAIN OF TOMATO AUCUBA MOSAIC VIRUS

## C. R. DAS AND S. P. RAYCHAUDHURI

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## INTRODUCTION

During the winter of 1949-50, tomato (Lycopersicon esculentum Mill.) crop grown in the experimental plots of Mycology and Botany Divisions at the Indian Agricultural Research Institute, New Delhi was found affected by a mosaic disease and the disease incidence varied from 2.7 to 4.5 per cent. The disease has been found to be readily transmissible by sap inoculation. Host range and properties of the causal virus have been studied with a view to establish its indentity and an account of the experimental work is reported briefly in this paper. Briant (1932), Berkeley (1937), Newton and Edward (1937), Friedeietsohn (1937), Clint (1941), and Wei and Cheo (1944) recorded the occurrence of a similar disease from Trinidad, Canada, Vancouver Island and British Columbia, U.S.S.R., Eire and China respectively.

## SYMPTOMS OF THE DISEASE

The earliest symptoms under field conditions were observed on young leaves in the form of mild mottling. In severe cases, however, almost the whole leaf surface became yellowish with scattered patches of green areas (Plate I, Fig. 1). The leaf surface appeared to be crinkled and brittle. Later, the whole leaf exhibited signs of withering and finally dropped.

Under glasshouse conditions the first signs of the disease appeared on the young leaves within 7 to 12 days after inoculation at 27-30°C. while the symptoms appeared after 3 weeks at 14-18°C. Chlorosis on leaves generally developed from the base but occasionally from the margins and the chlorotic areas rapidly increased and tended to coalesce. A tendency of masking of symptoms was observed during extreme cold in the winter season.

## MATERIAL AND METHODS

Tomato variety "Sutton's Early market" was used throughout and the cultures were raised and maintained in the insect proof house. The infected leaves were crushed and standard extract\* was used for all experimental work except where otherwise mentioned. Inoculations were made by gently rubbing the leaves with a sterilised swab of cotton wool previously dipped in the inoculum to

<sup>\*</sup>Standard extract w's prepared by adding 1 cc. of sterilized water to every gram of diseased leaf material.



Tomato leaves affected by Aucuba Mosaic



PLATE I

Symptoms of Aucuba Mosaic on *Nicotiana tabacum* var. White Burley.

Symptoms of Aucuba Mosaic on Solanum nigruin.

which carborundum powder had been added. The inoculated plants were kept under observation for a period of four weeks.

## EXPERIMENTAL

(a) Host-range:—In order to determine the host-range of the virus a number of plants belonging to different families were inoculated. The virus did not infect Capsicum frutescens L., Pisum sativum L., Crotalaria mucronata Desv., Arachis hypogaea L., Trifolium alexandrinum L., Abelmoschus esculentus (L.) Moench, Gossypium hirsutum L., Zinnia elegans Jacq. and Lagenaria siceraria Standl., but was readily transmitted to Solanum nigrum L., S. melongena L., Nicotiana tabacum L., N. tabacum var. White Burley, N. rustica L., Datura stramonium L., and Petunia sp.

## Symptomoatology:

Nicotiana tabacum:—Symptoms of the desease in the form of faint mosaic mottling which later became prominent appeard within a week of inoculation of plants at 27-30°C. but after about a month at 14-18°C. In some cases vein-banding was also observed. On N. tabacum var. White Burley vein-banding was however more prominent. (Plate I, Fig. 2A).

N. rustica: — Small circular faint chlorotic areas later turning necrotic appeared all over the lamina in about five days.

Sola num nigrum:—The symptoms developed within a month at 14-18°C. while at 27-30°C. these appeared in about 7 days. The first visible symptoms appeared generally at the base of the young leaves and spread in the form of mild mottling towards the apex. Later, interveinal mottling developed into a prominent symptom. (Plate I, Fig. 2B.).

S. melongena:—The symptoms appeared within nine days after inoculation as minute faint local lesions, which turned necortic.

Datura stramonium:—The disease in Datura appeared as small circular necrotic lesions on the inoculated leaves in nine days. These spots with whitish centre surrounded by pale areas tended to coalesce and form larger necrotic areas.

Petunia sp.:—The first obvious symptoms developed within five to six days as circular pinkish necrotic spots which increased in diameter with age resulting in drying up of the infected leaves.

(b) Properties of the virus:—(i) Thermal death point:—Thermal death point of the virus was determined by heating the same volume of standard extract in test tubes of equal diameter in a water bath at desired temperatures for 10 minutes. These heated virus extracts in test tubes were immediately dipped in cold water to bring down the temperature of the juice. The data given in Table 1 show that the virus withstood heating for 10 minutes at 98°C.

TABLE 1

Thermal inactivation of the virus

Exposure temperature	Number of plants inoculated	Number infected
40 50 60 70 80 90 95 98	8 8 8 8 8 14 14 14 6	4 4 2 3 3 7 4 4

(ii) Dilution end-point:—In order to determine the dilution end point of the virus the crude juice was diluted to desired strengths with distilled water. Inoculations were made on young and actively growing tomato plants. The results of this experiment are given in Table 2.

TABLE 2

Dilution end-point of the Virus

Dilution	Number of plants inoculated	Number infected
Crude sap 1: 10 1: 100 1: 1000 1: 1000 1: 5000 1: 10,000 1: 15,000 1: 20,000 1: 30,000 1: 40,000 1: 50,000 1: 100,000 1: 200,000	6 6 6 6 6 6 6 6 6 6 6	4 3 3 3 2 2 2 2 2 2 2 2

The data in Table 2 indicate that the virus tolerated a dilution of 1: 100,000 but not 1: 200,000.

- (iii) Longevity in vitro:—The standard extract was stored at laboratory temperature (18-30°C.) Young actively growing tomato plants were inoculated with this inoculum at intervals of 7 days and it was found that the virus remained active after storage for 77 days.
- (iv) Effect of desiccation on infectivity:—In order to determine whether or not the infectivity of the virus was retained in dry leaves, fully infected leaves showing prominent symptoms were collected and stored in a desiccator at room temperature (18-30°C). Young tomato plants were inoculated at intervals of 7 days with the dry material which was powdered and mixed with distilled water (for 1 gm. of dry leaf 10 cc. of water was added). The virus remained infectious even after 105 days in dried leaves.
- (v) Resistance to alcohol:—The standard extract of the virus was treated with ethyl alcohol at different concentrations in order to determine if the virus was inactivated by alcohol. Inoculations were made immediately on young leaves of tomato plants. This experiment indicated that the virus remained active when treated with 90% alcohol but lost its infectivity when treated with 95% alcohol.

## DISCUSSION

The data indicate that some of the properties e. g. dilution endpoint and resistance to alcohol correspond with the tomato aucuba mosaic virus as described by Smith (1928) and Nakata and Takimoto (1940), while it differs in thermal death point. Bewley and Corbett (1928) reported that tomato ancuba mosaic produced stripe symptom on Solanum melongena. The virus under study, however, showed local necrotic lesions on the inoculated leaves of eggplants. Caldwell (1932) reported that on Solanum nigrum and Nicotiana tabacum including the variety White Burley, it produced mottling on leaves, while on Datura stramonium it developed local necrotic spots. In Nicotiana tabacum including the variety White Burley, the virus induced mottling at the initial stage of infection and later vein-banding developed while on Solanum nigrum the virus produced interveinal mottling. The symptom on Datura stramonium appeared to be similar to that described by Caldwell (1932). Kunkel (1932) observed that tomato aucuba mosaic virus produced necrotic local lesions on the leaves of N. rustica.

Allard (1915) stated that in tobacco mosaic virus attenuation is indicated at the dilution of 1:10,000. McKinney (1927) observed that the temperature at which the virus of tobocco mosaic becomes inactivated depends on the concentration of the virus and the nature of the plant juice. Kassanis and Salman (1947) reported that tomato aucuba mosaic virus maintained in Kondine Red tomato plants continually developed necrotic local lesion symptom when transmitted to White Burley tobacco, but a different result was obtained afterwards. Instead of necrotic local lesions it produced systemic infection showing mosaic symptom. It was found that tobacco variety White Burley existed in two distinct lines, one developing necrotic local lesions while the other developing mottling with the same

inoculum. Kunkel (1934) observed three distinct strains of aucuba mosaic virus from plants incubated at high temperature for varying periods after inoculation. A number of other forms of aucuba mosaic appeared to be intermediate which he referred to as attenuated strains.

While most of the properties of the virus under study are similar to tomato aucuba mosaic virus, there are others in which it differs. It therefore, appears to be a strain of tomato aucuba mosaic.

## SUMMARY

A mosaic disease of Lycopersicon esculentum characterised by interveinal mottling and yellowing is described. The virus is readilly transmissible by sap inoculation. The virus transmissible to Solanum nigrum, S. melongena, N. rustica, Datura stramonium and Petunia sp. withstands temperature of 98°C. and dilution of 1:100,000. Longevity in vitro is more than 77 days and the virus retains its infectivity after treatment with 90% alcohol but is inactivated by 95% alcohol. The virus remains active even after 105 days in dried tissue at room temperature (18–30°C.).

The causal virus appears to be a strain of tomato aucuba mosaic virus.

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# OCCURRENCE OF POWDERY MILDEW OF WHEAT IN THE NEIGHBOURHOOD OF JODHPUR

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## INTRODUCTION

The area under wheat in the neighbourhood of Jodhpur is significantly small being limited to the low lying tracts in between rocky hillocks. The general climate of the area during winter is dry with occasional light showers. The maximum temperature reaches 100°F during March and the minimum falls to 40°F during December and January. During rainy season, water gets collected in these low lying lands and remains so till November and in some cases till December. Water logging often delays the sowing of wheat and also helps in maintaining soil humidity. Irrigation is carried out through wells.

Every year, wheat is attacked by the powdery mildew, caused by Erysiphe graminis tritici El. Marchal. In the month of March 1952, heavy damage to the wheat crop due to this disease was observed.

The oidial stage was abundant; the cleistothecial stage, although not so common was found when the crop was maturing. In the cleistothecium, asci were invariably present but no ascospore formation was observed. Factors governing the preparation of the disease were, therefore, studied.

## MATERIAL AND METHODS

Artificial inoculations of wheat plants were carried out by smearing the leaf surface with the conidial material. Inoculated plants were kept under saturated humid conditions for 48 hours. Before making inoculations, conidial material, obtained from the field or artificially inoculated plants, was subjected to freezing conditions for about half an hour as suggested by Graf-Marine (1934). Every time the inoculations were made, germinability of conidia was tested.

For all pathological studies, the local wheat variety, on which the disease appears, has been used. Other wheat varieties which have been used for testing their reactions to the mildew were obtained from the Division of Botany, Indian Agricultural Research Institute, New Delhi.

Experiments about germination and pathological studies were repeated thrice to obtain convincing results.

For germination studies, the method suggested by Thirumalachar (1940) was used.

Wheat leaf extracts were prepared by boiling 250 grams of green wheat leaves in 1,000 c.c. of distilled water. The extract was filtered through a muslin cloth and autoclaved at 15 lb. pressure for 20 minutes.

## PHYSIOLOGICAL STUDIES

Following factors effecting formation of ascospores in cleistothecia have been studied:--

- 1. Effect of temperature:—The cleistothecia were subjected to temperatures of  $0^{\circ}$ ,  $5^{\circ}$  and  $9^{\circ}C$  for different intervals of time. The material was afterwards exposed to a room temperature of  $21^{\circ}\pm1^{\circ}C$  and observations were made at different intervals of time. Positive results of ascospore formation were obtained with the material frozen at  $5^{\circ}$  and  $9^{\circ}C$  for 8 hours after an exposure at room temperature for 48 and 24 hours respectively. (Fig. 1)
- 2. Effect of sucrose concentrations:—The cleistothecia were subjected to sucrose concentrations ranging from 2 to 30 per cent. Positive results, with clear ascospore formation were obtained at 2, 10 and 25 per cent after an exposure for 120, 24 and 17 hours respectively. (Fig. 2)
- 3. Effect of Nitric acid concentrations:—Different concentrations of the acid were prepared by adding different number of standard pippette drops of the concentrated Nitric acid to 5 c.c. of sterilized water. The cleistothecia were subjected to these acid dilutions for different periods. Positive results were obtained at a concentration of 8 pippette drops in 5 c.c. of water after an exposure of 144 hours. (Fig. 3)
- 4. Effct of Potassium Nitrate concentrations:—The cleistothecia were subjected to different concentrations of Potassium nitrate solution ranging from 2 to 30 per cent for different periods. The positive results, by mere segmentations of cytoplasmic contents, were obtained with 25 per cent Potassium nitrate solution after 168 hours. (Fig. 4).

No positive results of ascospore formation could be obtained when the material was subjected to (i) intermittent wetting and drying, (ii) different concentrations of vitamins Thiamine, riboflavin and nicotinic acid and (iii) different soil conditions analogous to those found in nature.

## GERMINATION STUDIES

1. Effect of Relative humidity:—To obtain atmosphere with different percentages of moisture, Mclean and Cook's (1941) method, using different dilutions of sulpuric acid, has been employed. For humidity chambers, tightly fit petriplates were used. Dry clean slides were dusted with conidia and immediately placed in the humid chambers at 25°C.

Slides were examined after 4, 9, 20, 30 and 36 hours. The results obtained show that the time required for the conidial germination is the least and the percentage of germination together with the

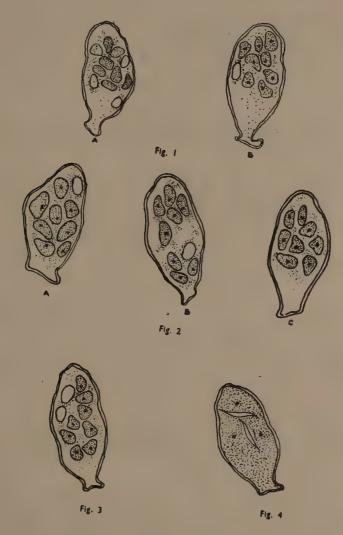


Fig. 1. Ascospore formation at different temperatures of freezing.

A. At 5°C. B. At 9°C.

Fig. 2. Ascospore formation in sucrose solutions.

- A. In 25% sucrose sol. after 17 hours.
  B. In 10% sucrose sol. after 24 hours.
  C. In 2% sucrose sol. after 120 hours.

Fig. 3. Ascospore formation in nitric acid concentrations. (8 drops of HNO3 in 5c.c. of water) after 144 hours.

Fig. 4. Ascospore formation in 25% Potassium nitrate solution after 168 hours.

elongation of germ tubes highest (54 percent) at 100 percent relative humidity. However, the conidial germination is greatly handicapped in the actual drop of water where only 3 percent germination was obtained after an exposure of 30 hours. Another important observation made was that immediately after germination the germ tubes in the drop of water were assuming erect positions away from the drop. No germination was obtained below 92.9 percent relative humidity.

2. Effect of temperture:- Fresh viable conidia were kept for germination on glass slides in moist chambers maintaining 100 percent relative humidity at different temperatures. The slides were examined at intervals.

The results obtained indicate that the percentage of germination at 4° and 9°C is highest (61 & 63 percent respectively) but the progress of the germ tube is very slow (2.9–8.6 $\mu$ ). The percentage of germination at 25°C. is 52 but the length of germ tube is greatest (14.5–54.9 $\mu$ ). The fungus retains its germinability up-to 32°C. No germination was obtained at 34°C.

The results compare favorably with those obtained by Graf-Marin (1934) working with *Erysiphe graminis hordei* El. Marchal.

- 3. Effect of different nutrient media on germination:— The following media were used:
  - i. Extract of healthy wheat leaves.
  - ii. Extract of heavily mildewed wheat leaves.
  - iii. Czapek solution.
  - iv. 2.5 and 5.0 percent sucrose solutions.
  - v. Distilled water.

Czapek solution was perepared as suggested by Mclean and Cook (1941). Conidial suspensions were made from conidia of wheat leaves. The slides, placed in moist chambers, were kept at a constant temperature of 25°C. for 36 hours.

The results obtained, showed that at 2.5 percent sucrose solution, the germination percentage was highest. (40 percent) In the case of extract of healthy wheat leaves and czapek solution, the percentages of germination were 24 and 23 respectively, 5.0 percent sucrose solution gave only 8 percent germination. With distilled water, used as a control experiment, the germination was 3 percent. The extract of heavily mildewed leaves inhibited germination, in which case germination was only 1 percent.

Graf Marin (1934) found similar results in the case of Erysiphe graminis hordei El. Marchal.

4. Effect of leaf extracts of different wheat varieties on conidial germination:- Healthy leaf extracts of wheat varieties N. P. 4, N. P. 52, C.591 and local varieties were prepared. Conidial suspensions were made in these extracts and a drop of it put on the

slides for germination in moist chambers at 25°C. The results obtained indicate that the extracts of wheat varieties N.P. 4, N.P. 52 and C.591 do not show any significant adverse effect on the germination of conidia in comparision to that of the local wheat variety.

## PATHOLOGICAL STUDIES

1. Effect of temperature:- The influence of temperature on the intensity of infection was studied by making artificial inoculations, at intervals, during March and April 1952. Observations were made in terms of positive and negative reactions.

The results obtained show that the infection appeared with the inoculations made on 10th and 30th of March 1952 when the temperature varied between 69° to 95°F. and 76° to 100°F. respectively. No infection was obtained with the inoculation made on 20th April, when the temperature was between 78° and 104°F.

- Effect of moisture:- Inoculations with conidial material were made on wheat plants and the inoculated plants were kept under saturated humid conditions for different intervals of time. The results obtained showed that a minimum of 6 hours saturated humid condition was necessary for obtaining infection.
- Host range of fungus: Various members of the Gramineae and some other plants, such as Tephrosia purpurea, Ocimum sanctum, Coccinia indica, Melilotus alba, usually found to be infected with powdery mildews in nature, having more or less similar measurements of their conidia, were inoculated with the conidial material of Erysiphe graminis tritici El. Marchal. In no case infection was obtained showing that the fungus, under study, is highly specialized.
- Infection tests of some other powdery mildews upon wheat .:-Wheat plants were inoculated with conidia of different powdery mildews, collected in the neighbourhood of Jodhpur, but in no case postive results were obtained.

## INCIDENCE OF THE DISEASE

Corp and plant infections were recorded every fortnight in the months of March and April 1952. For 'crop-infection', the method followed was to count the number of healthy and infected plants in one square yard of space chosen at random, Five such random samples were taken and counts were made. Average of these 5 counts has been taken to represent the 'crop-infection' on that date.

To determine the 'plant-infection', counts of healthy and infected leaves on individual plants, chosen again at random, were made. Here 100 such plants were observed and the average of these counts represents the 'plant-infection' for that date.

These results, recorded in the table 1 show that the disease is not serious in the first week of its appearance but after about a week, it spreads rapidly and takes up severe proportions.

TABLE 1

Periodical Assessment of the Percentage of the Crop and Plant Infections

Date	Percentage of Crop- infection	Percentage of Plant- infection
10-3-52	28.7	5.3
25-3-52	61.3	18.1
9-4-52	86.1	23.8
24-4-52	97.4	26.2

## VIABILITY OF THE FUNGUS

In order to determine the viability of the fungus during summer months, following experiments were performed:—

- 1. The conidial material was subjected to different high temperatures for different periods in an incubator. After the treatment, the material was tested for the germinability of the conidia at 25°C. The results obtained indicate that the conidia lose their viability at 35° and 38°C after an exposure of 2 hours and 1 hour respectively. At 41°C, the viability was lost within half an hour.
- 2. The conidial material was kept from April to November 1952 under laboratory conditions. The material was tested for its germinability in the months of November 1952. It was found that the conidia showed no germination.
- 3. Heavily infected ear heads were kept under laboratory conditions from April to November 1952. This seed was sown in the month of November in sterlized soil. The pots were placed in glass cages to prevent outside contamination. It was found that the plants remained healthy showing no sign of infection.

## DISCUSSION

The role of cleistothecium in the annual recurrence of the disease has been a subject of much controversy. Brooks (1928) and Mehta (1930) could not find any role of cleistothecium in the annual recurrence of the disease. However, Foster and Henry (1937) reported that the fungus overwinters in the cleistothecial stage in Alberta.

During the present studies, weekly collections of the cleistothecial material were made and observed for ascospore production but it resulted in failure. The same material was subjected to different conditions analogous to those found in nature but again no positive results were obtained. However, ascoposre-formation was obtained by subjecting the material to different artificial conditions, not available in nature. Therefore there remains practically no chance to suggest the positive role of cleistothecia in conditions of the area under investigation,

During these studies, it was considered that possibly Mt. Abu might be playing an important role in the annual recurrence of the disease. Mt. Abu was, therefore, visited twice on 25 October and 14 December 1952. The wheat fields were visited and searched vigorously for the disease. The plant debris was also collected from those fields having wheat crop in the previous season and lying fallow afterwards, and observed. But both these attempts resulted in failure to give any clue to establish that Mt. Abu plays any role in the recurrence of the disease.

Mehta (1930) concluded that the annual recurrence at the foot of hills and plains is through wind-blown conidía from hills in the Himalayas. There remains a strong possibility of the Himalayas forming the source of recurrence.

## SUMMARY

The development of asci in cleistothecia under the conditions is quite frequent but no ascospore formation could be observed.

Effects of temperatures, sucrose-concentrations, nitric acid concentrations, potassium nitrate concentrations, intermittent wetting and drying, vitamins and different soil-conditions on the ascospore formation have been studied. Positive results are obtained in the case of temperature, sucrose-concentrations, nitric acid concentrations and potassium nitrate concentrations.

Effect of relative humidities, temperature, nutritive media and leaf-extracts of different varieties of wheat on the germination of conidia have been studied.

Temperature and moisture have been found to have a profound effect on infection. No infection was obtained when the prevailing minimum and maximum temperatures were 78° F and 104° F respectively. It has been observed that a minimum of six hours of saturated humid conditions is necessary for obtaining infection. It has been found that the fungus is strictly specialized in its host-range.

Observations on the incidence of the disease in the field have indicated that the disease is most severe from the 1st week of March onwards.

Viability of the fungus, tested under different conditions has shown that the fungus loses its viability during the period from May onwards.

Annual recurrence of the disease has been discussed and in the light of the results obtained, it has been co-concluded that the cleistothecia are probably functionless and every year the infection is through conidia which are wind-blown.

The authors express their sincere gratitude to Prof. Shanti Sarup, Head of the Botany Department, Jaswant College, Jodhpur, for his constructive criticism and encouragement given and the facilities provided throughout the course of this investigation. We are

specially thankful to the Director, Indian Agricultural Research Institute, New Delhi, for having given us the facilities for consulting the library and to Head of the Division of Botany for supplying some wheat varieties.

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## A REVIEW OF BACTERIAL PLANT DISEASE INVESTIGATION IN INDIA

M. K. PATEL AND Y. S. KULKARNI

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Smith (1920) stated in his "Introduction to Bacterial Plant Diseases" that Asia is "terra incognita" so far as the knowledge of bacterial plant pathogens was considered. That statement could not be considered precise, for, by the time that book was published, at least four papers reporting the occurrence of bacterial plant diseases had appeared in India, an Asian country. Indeed, hardly had the controversy between Smith and Fischer about the occurrence of bacterial diseases in plants subsided, when Butler (1903) reported the occurrence of "bangle blight" of potatoes caused by Phytobacterium solanacearum (Smith) Patel and Kulkarni in this country.

Even though the reports on the occurrence of bacterial plant diseases in India were few, the diseases must have been present for a long time. Fawcett (1936) found lesions of citrus canker due to Xanthomonas citri (Hasse) Dowson on herbarium specimens of citrus plants at the Kew herbarium collected somewhere in Eastern India between 1827 and 1831. In the specimens of mango leaves, collected by Johnston in Bihar in 1881, by Inayatulla Khan in Kheri and by Gamble at Dehra Dun and deposited in the herbarium at Dehra Dun, lesions similar to those caused by Phytobact. magniferae indicae (Patel, Moniz and Kulkarni) Patel and Kulkarni at Poona have been noted by the Forest Botanist (Personal communication). Critical examination of other herbarium specimens in the various herbaria, of plants collected in the past, is likely to reveal a similar state of affairs.

Butler (1903) considered the "bangle blight" of potatoes to be similar to and identical with Smith's "brown rot" of potatoes. Coleman (1909), Mann and Nagpurkar (1920) and Mitra (1937) recorded this disease in Mysore and Bombay States respectively. Since then the disease has been found to be of wide distribution in causing much damage to the potato crop.

Further investigations on bacterial plant diseases were made, many of them as a result of the impetus given by the work of Smith himself. Hutchinson (1913) investigated a wilt of tobacco in Rangpur district in Bengal and identified the causal organism as *Phytobact*. solanacearum though it was unable to cause wilt in potato and tomato plants. Another disease investigated by Hutchinson (1917) and Chaudhari (1935) was a bacterial disease of the ears of wheat, locally known as "tundu" disease caused by Aplanobacter tritici (Hutchinson) Burkholder. It appears to be a doubtful species of the

TABLE 1

List of bacterial plant pathogens reported from the Plant Pathological Laboratory, Poona.

												natus,		
Hosts	Desmodium diffusum	Desmodium diffusum-gangeticum	Ipomoea muricata	Xanthium strumarium and Pisum sativum	Cajanus cajan	Cassia tora and Pisum sativum	Tamarindus indica and Caesalpinia sepiaria	Lawsonia alba	Sesbania aegyptiaca	Clerodendron spp.	Piper betle	Phaseolus vulgaris var. White-Kidney, P. lunatus, P. coccineus and Dolichos lablab	Euphorbia pulcherrima	Stizolobium deeringianum
Organism	Xanthomonas desmodii Uppal and Patel*	X.desmodii-gangeticii Uppal, Patel and Moniz*	X.uppalii Patel*	X.badrii Patel, Kulkarni and Dhande (1950)	X.cajamii Kulkarni, Patel and Abhyankar (1950)	X.cassiae Kulkarni, Patel and Dhande (1951)	X.tamarindi Patel, Bhatt and Kulkarni (1951)	X.lawsoniae Patel, Bhatt and Kulkarni (1951)	X.sesbaniae Patel, Kulkarni and Dhande (1952)	X.clerodendroni Patel, Kulkarni and Dhande (1952)	X.betlicola Patel, Kulkarni and Dhande (1951)	X.phaseoli (Smith) Dowson var. indicus Uppal, Patel and Nikam*	X.poinsettiaecola Patel, Bhatt and Kulkarni (1951)	X.stizolobiicola Patel, Kulkarni and Dhande (1951)
S.No.	-	2	co	4	5	9	7	00	6	10	Ħ	12	13	14

Medicago sativa, Pisum sativum, Trigonella foenum-graecum and Melilotus indica	Begonia spp.	Brassica spp. and Raphanus sativus	Gossypium spp.	Glycine max	Ricinus communis	Capsicum annuum and unripe tomato fruits	Vigna sinensis, V. sesquipedalis and Phaseolus vulgaris	Citrus aurantifolia and other Citrus spp.	Solanum tuberosum and Lycopersicum esculentum	Mangifera indica	Vitis woodrowii Ripe mango fruit and vegetables
X.alfalfae Riker et al (see Patel, Kulkarni and Dhande (1949)	X.begoniae (Takimoto) Dowson (see Patel, Kulkarni Begonia spp. and Dhande (1951)	X.campestris (Pammel) Dowson*	X.malvacearum (Smith) Dowson*	X.phaseoli-sojense (Hedges) Dowson*	X.ricinicola (Elliott) Dowson (see Patel, Kulkarni and Dhande 1951)	X.vesicatoria (Doidge) Dowson (see Patel, Kulkatni Capsicum annuum and unripe tomato fruits and Dhande (1950)	X. vignicola Burkholder*	X. citri (Hasse) Dowson*	Phytobacterium solanacearum (Sm.) Patel and Kulkarni (1952)	P.mangiferae indicae (Patel, Moniz and Kulkarni) Patel and Kulkarni*	P.vitis-woodrowii Patel and Kulkarni (1951) Pectobacterium carotovorum (Jones) Waldee (see Patel and Padhye (1948)
15	16	17	18	19	20	21	22	23	24	25	27

\*These have been referred to by Patel, Abhyankar and Kulkarni (1948).

genus Aplanobactor as it has never been proved pathogenic by itself. Ballard and Norris (1923) observed that the 'angular leaf-spot' disease of cotton incited by X.malvacearum (Sm.) Dowson was prevalent in parts of Madras State. Black rot of cabbage due to X.campestris (Pammel) Dowson might have been present in India for a long time, but the first authentic report of its occurrance was made by Patwardhan (1928) from Poona and has subsequently been reported from most parts of the Indian Union. A becterial soft-rot of garden poppies (Papaver somniferum and P. rhoeas) caused by Pectobact. aroideae (Townsend) Waldee (=Erwinia papaveris (Ayyar) Magrou) was noticed by Ayyar (1927) at Kanpur, Pusa. Four years later, Prasad (1930) isolated the same organism from turnip suffering from Aerobacter dissolvens (Rosen) Waldee, reported by Prasad (1930) as causing a soft-rot of maize, has now been proved to be a saprophyte. Prasad (1930) also reported a bacterial leaf-spot of cucumber (Cucumis sativus) due to X. cucurbitae (Bryan) Dowson. The two organisms viz., Pseudomonas desaiana Burkholder (=Bacterium)pyocyaneus-saccharum Desai) and Bacillus fructodestruens Madhok and Fazal, causing rots sugarcane tops and tomato fruits respectively, as reported by Desai (1935) and Madhok and Fazal-ud-din (1942), seem to be of doubtful validity as they are not stated to be pathogenic on living hosts. McRae (1933) and Padwick (1940) report occurrance of X. rubrilineans (Lee et al) Starr and Burkholder on Saccharum officinarum. Asthana and Mahmud (1944) report bacterial leaf-spot of Piper betle in Madhya Pradesh. Singh (1943) recorded the possible occurrance of "hairy root" due to Agrobacterium rhizogenes (Riker et al) Conn, and crown gall due to Agrobact. tumefaciens (Sm.) Conn and fire blight of apple due to Erwinia amylovora in Uttar Pradesh. It is not known whether these were introduced along with the imported root-stocks or through some other source and to what extent the diseases have got established.

Since 1947, a number of papers on bacterial diseases of fruit, forest and leguminous crops and weeds have been published by Patel and his associates. At present, 23 organisms belonging to the genus Xanthomonas, 3 to Phytobacterium and one to Pectobacterium have been reported from the Plant Pathological Laboratory at Poona. Nine of these, although described earlier by other workers, are reported for the first time in India and 17 are proposed as new species, details about which are recorded in table I.

From table 1 it is clear that the organisms X. desmodii, X. desmodii-gangeticii, X. uppalii, X. badrii, X. cajani, X. cassiae, X. tamarindi, X. lawsoniae, X. sesbaniae and X. clerodendroni are found on plants indigenous to India while X. betlicola, X. stizolobiicola and X. vignicola, although described earlier by foreign workers, are new records for India. It is not certain whether the two organisms X. cucurbitae (Bryan) Dowson on cucumber and X. ricinicola (Elliott) Dowson on castor were originally present in India or introduced into this country since the hosts on which these occur are extensively cultivated and the diseases on them are fairly widespread. There are, however, strong reasons to believe that *Phytobact*. solanacearum, Pectobacterium carotovorum, X. campestris, X. phaseoli-sojense, X.

alfalfae, X. begoniae and X. poinsettiaecola gained entry into this country from the West on seeds and stocks as India had depended largely upon foreign countries for potato, cabbage, soyabean and lucerne seeds as well as ornamental plants such as begonia and poinsettia. Even so, X. vesicatoria may have got introduced into our country along with imported seeds of chillie or tomato from America where it incites a severe disease on these hosts. Recently, Hedayetullah and Saha (1941) and Jain (1951) have reported a bacterial canker of tomato fruits due to Aplanobacter michiganense Sm. Fire blight of Cosmos bipinnatus caused by Erwinia cosmovora has been recently recorded by Prasad (1952) at Anand (Dist. Kaira). Patel and Padhye (1948) and Hingorani (1951) recorded Erwinia carotovora causing soft rot of vegetables. Hingorani and Malla (1951) reported Ps. marginalis causing soft rot of onions in storage.

Besides X. citri, Phytobact. mangiferae-indicae seems probably to be another orgnism to have left the shores of this country on mango grafts, as the disease caused by it in India is quite similar to that described by Doidge (1915) in Africa.

Knight and Hutchinson (1950) are of the opinion that 'blackarm' is a disease of the 'Old World' cottons (of Indian origin) and was introduced into U. S. A. According to Knight (1948), most of the Indian cottons, both Gossypium arboreum and G. herbaceum, show universal immunity to the disease under influence of long and intense selection by the disease. Patel and Kulkarni (1950) and Balasubrahmanyan and Iyengar (1952) on the other hand find that varieties of both G. arboreum and G. herbaceum are susceptible to blackarm. The difference in the reactions as reported by Knight (1948) and Patel and Kulkarni (1950) may be due to different environmental conditions under which the tests for resistance were carried out.

Of the 27 bacterial diseases mentioned in table 1, only 5, viz. X. citri, X. campestris, X. phaseoli var. indicus, X. malvacearum and Phytobact. solanacearum may be considered as of major importance as they cause serious damage to crops.

Citrus canker is well established throughout the length and breadth of India. Pruning followed by spraying with Bordeaux mixture and a suitable inseticide has proved a promising control measure. Disinfecting cabbage seeds in mercuric bichloride solution (1:1000) for 30 minutes has also given encouraging results in that the seedlings raised from such seeds are completely free from initial infection of black rot. Blackarm of cotton is more serious on exotic cotton varieties (Gossypium hirsutum), especially in continuous wet seasons. Attempts are being directed towards isolating varieties highly resistant to blackarm under artificial epidemics in the field. A few indigenous cotton varieties such as K. F. (G. herbaceum var. acerifolium) and Red arboreum (G. arboreum race bengalense) show a very high degree of resistance to blackarm. Recently, Kulkarni and Patel (1951) have shown that resistance to blackarm in G. herbaceum is governed by a single gene, susceptibility being partially dominant. According to Patel. Kulkarni and Kulkarni (1952), the problem of ring disease of

potatoes is practically solved in Bombay State by planting fresh healthy seed brought from disease-free areas in Simla Hills.

In table 2 are recorded 36 genera of Indian plants (comprising 17 families) within the limits of which one or more species are subject to bacterial diseases.

TABLE 2

Showing families and genera, some species of which are subject to bacterial diseases as reported from this laboratory.

S. No.	Families	Genera
1 :	Anacardiaceae	Mangifera
$\hat{\mathbf{z}}$	Begoniaceae	Begonia
$\overline{3}$	Compositae	Xanthium
3	Convolvulaceae	Ipomoea .
5	Cruciferae ·	(1) Brassica
		(2) Lepidium
		(3) Raphanus
6	Cucurbitaceae	Cucumis
7	Euphorbiaceae	(1) Euphorbia
	•	(2) Ricinus
8	Leguminosae	(1) Caesalpinia
		(2) Cajanus
		(3) Cassia
		(4) Desmodium
t		(5) Dolichos
!		(6) Glycine
		(7) Medicago
1 .		(8) Melilotus
		(9) Phaseolus
		(10) Pisum
•		(11) Tamarindus
		(12) Trigonella (13) Sesbania
		(14) Stizolobium
		(15) Vigna
9	Lythraceae	Lawsonia
10	Malvaceae	Gossypium
11 .	Papaveraceae	Papaver
12	Piperaceae	Piper
13	Rosaceae	Pyrus
14	Rutaceae	Citrus
15	Solanaceae	(1) Capsicum
1		(2) Lycopersicum
		(2) Lycopersicum (3) Solanum
16	Verbenaceae	Clerodendron
17	Vitaceae	Vitis

In table 3 is given a list of 12 families and 18 genera, some species of which are suspected to be attacked by one or more bacterial diseases.

TABLE 3

Families and genera, some species of which are suspected to be attacked by one or more bacterial diseases.

S. No.	Families	Genera
1 2 3 4 5 6	Amaranthaceae Liliaceae Boragineae Compositae Geraniaceae Gramineae	Amaranthus Allium Trichodesma Zinnia Geranium (1) Eleusine (2) Oryza (3) Sorghum
<b>7</b>	Iridaceae Leguminosae	(4) Triticum Gladiolus (1) Alysicarpus (2) Erythrina (3) Rothia
9 10 11 12	Moraceae Pedaliaceae Solanaceae Verbenaceae	(4) Butea Morus Sesamum Nicotiana Tectona

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#### PHYTOPATHOLOGICAL NOTES

A new Physiologic Race of Puccinia graminis tritici (Pers.) Erikss and Henn. in India. R. S. Vasudeva. V. C. Lele and D. P. Misra. A new physiologic race of black rust of wheat, not hitherto recorded from India\*, has been picked up from the South from the Rabi crop of 1952-53. The collection contained, besides new race, another race 21 which has been most common in recent years. Types of infection produced by the new race of Puccinia graminis tritici on the differential hosts are given below:—

Stock collection or isolation	Little club	Marquis	Reliance	Kota	Arnautka	Mindum	Spelmar	Kubanka	Acme	Einkorn	Vernal	Khapli
Coimbatore Kenphad 28 Arn (2)	×	0:-2	0;	:0	0;-2	0;-2	0;-2	4	4	3-4	0; -2	4

This race is new and different from all other known Indian races in that Arnautka, Mindum and Spelmar show resistance to it. Little club shows "type X" infection with this race, showing high degree of resistance at higher temperatures (49–88°F) and moderate susceptibility at lower temperatures (40–79°F).

The new race is allied to race 72, also not reported from India and differs from it but slightly.—Division of Mycology and Plant Pathology, I. A. R. I., New Delhi.

Saponaria Leaf Curl. R. N. Azad. A severe leaf-curl disease was observed during 1952 at Delhi in Saponaria vaccaria L., which is a popular flowering winter annual. It was so severe that the Saponaria beds were almost devastated by the disease. The disease was again observed during January, 1953.

The diseased plants are stunted in growth; the leaves are curled and greatly reduced in size. Profuse, irregular, granular out-growths appear on the veins on the under-side of the leaves which is a characteristic symptom of the disease. The flowering in the diseased plants is either scanty or totally absent. In case of early infection i. e.

<sup>\*</sup>Mehta, K. C. (1940) Further Studies on Cereal Rusts in India I. C. A. R., Scin.

Monograph No. 14.

Prasada, R. & V. C. Lele New physiologic races of wheat rusts in India, Indian (1952) Phytopathology 5: 128-129 (Issued 1953).

Uppal, B. N. and A new race of Puccinia graminis tritici and two biotypes V. P. Gokhale, (1947) of race 42. Curr. Sci. XVI, 2. p. 61.



Fig. 1:— A plant of Saponaria vaccaria infected in seedling stage.



Fig. 2:—Transmission of the disease by grafting.

when infection takes place in seedling stage, the plant does not grow more than 5-6 inches in height with few large leaves and numerous small, curled leaves condensed on the stem. During later stages the leaves sometimes become purple. Fig. 1 shows a plant infected in seedling stage.

The disease was successfully transmitted to healthy plants of Saponaria vaccaria by grafting and cent per cent infection was obtained. After about a month of grafting the new growth on the stock developed the typical symptoms of the disease. The transmission studies were carried out in an insect-proof glass-house. Fig. 2 shows transmission of the disease by grafting.

All efforts to transmit the disease to Saponaria vaccaria or any other plant species by sap inoculation remained unsuccessful. As it was suspected that the disease in Saponaria vaccaria might be due to tobacco leaf-curl virus (Nicotiana virus 10), which causes almost similar symptoms on many hosts in India, transmission tests with white flies (Bemisia tabaci Gen.), the vector of Nicotiana virus 10, were repeatedly made on Saponaria vaccaria, tobacco and tomato. The white flies failed to transmit the disease to any host. The results show that the virus causing leaf-curl in Saponaria vaccaria is different from the tobacco leaf-curl virus (Nicotiana virus 10).

This is the first record of a virus disease in Saponaria vaccaria L. Division of Mycology & Plant Pathology, Indian Agricultural Research Institute, New Delhi.

Physalis Peruviana L. a new host of tobacco leaf-curl virus. T. K. Nariani and P. S. Pathanian. During the summer of 1953 a severe leaf-curl disease of Physalis peruviana L. was observed at the Indian Agricultural Research Institute. The diseased plants are severely stunted in growth and are characterised by curling, puckering and smalling of the leaves and presence of dark green enations on the veins on their under surface (Fig. 1). The veins and veinlets are very prominent and thickened. The enations are oval-or cupshaped, frilled and sessile. In severe cases the margins of the leaves are curled upwards forming cup like structures but in mild cases curling is only slight but the enations are always present.

The disease was readily transmitted within 2-3 weeks when diseased shoots of *P. peruviana* were grafted to healthy plants raised through seed in the insect-proof house. The axillary shoots growing from the stock below the grafted portion showed severe leaf-curl with enations. The disease could not, however, be transmitted when juice extracted from the leaves of diseased plants was rubbed with carborundum powder on healthy plants of *P. peruviana*.

Insect transmission tests were conducted with white fly (Bemisia tabaci Gen.). About 8-15 white flies previously fed for 16 hours on diseased plants were liberated on 7 healthy plants of P. peruviana for 24-48 hours. Typical symptoms of the disease including enations appeared within 15-25 days on all the plants.



Fig. 1:—Leaves of diseased *Physalis peruviana* L. showing enations on the veins on the undersurface.



Fig. 2:—Transmission of the virus to tobacco by grafting producing tobacco leaf curl.

The disease was also transmitted to *Nicotiana tabacum* var. Harrisons' Special by grafting (Fig. 2) as well as by the agency of white fly. All the six plants of tobacco fed with viruliferous white flies produced typical curling, puckering and cup-shaped enations characterestic of tobacco leaf-curl virus (*Nicotiana* virus 10) showing thereby that the causal virus is the same as that of tobacco leaf-curl.

Grateful thanks are due to Dr. R. S. Vasudeva, Head of the Division of Mycology and Plant Pathology for guidance and helpful suggestions throughout this investigation.—Division of Mycology and Plant Pathology, Indian Agricultural Research Institute, New Delhi.

### **ANNOUNCEMENT**

### INDIAN PHYTOPATHOLOGICAL SOCIETY

Pusa Buildings, New Delhi-12.

July, 22 1951.

Dear Colleague,

It has been proposed by several members of the Indian Phytopathological Society that a half-day session should be arranged during the next Annual General Meeting of the Indian Phytopathological Society to be held in January, 1955, at Baroda during the Science Congress week. Besides the Presidential address and election of new office-bearers and other routine business, papers of Phytopathological interest may be read or a symposium on certain aspect of plant disease may be held if there be a good response from the members attending the next Annual Meeting at Baroda. I would, therefore, be obliged if you will kindly let me know by 28th August, 1954, whether you are likely to attend the meeting and participate in the proposed session by presenting papers for reading and discussion. Any other suggestions would be most welcome.

Yours sincerely, R. PRASADA, Secretary.

### Editorial Board for 1954

For Volume VII, 1954, the Councillors have elected the following members to form the Editorial Board:

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The members are requested to send their articles intended for publication to the undersigned who will forward them to the Chief Editor. This arrangement has been found most suitable in view of the fact that the Editorial Board is elected only for one year, and the press is in New Delhi.

The Editorial Board for 1954 was not announced earlier because it would have served no useful purpose when 1953 numbers had not been issued.

R. PRASADA
Secretary-Treasurer.

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Contributions should be on one side of the page, double spaced, with a 1-1/4th inch margin on the left. In form and style, such as punctuation, spelling and use of italics, the manuscript should conform to the best Journals in the U.K. and U.S.A. Authors should strive for a clear and concise style of writing. The name and address of the Institution at which the work was done should be cited immediately after the SUMMARY at the end of the article on left hand side. Tables should be numbered and each table should have a heading stating briefly its c ntents. References to literature should be made as foot notes only when four or fewer citations are given. If there are more, they should be listed under 'REFERENCES' at the end of the paper and referred to by date in brackets in the body of the article. Citation should give the name of the author (or others), his (or their) initials year of publication and then the full title correctly, followed by the name of the Journals, number of the volume, a colon and page numbers. If the title is in a foreign language, then diacretic signs and capitalization should be precisely as in the original. The names of the Journal should be as abbreviated in the WORLD LIST OF PERIODICALS, 2nd Ed., 1934, but as that book may not be available to all, contributors are requested to give the titles in full. Abbreviating will in that case, be done by the Editors. If an article has not been seen in original, then that fact should be clearly stated. An example citing is given below:-

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Authors are invited to consult Bisby's 'An Introduction to Taxonomy and Nomenclature of Fungi' (1945), pp. 38-41 and Riker's 'The preparation of manuscripts for *Phytopathology*, *Phytopathology* 36: 953-977, 1946, before preparing their mss. and figures.

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